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(12) **United States Patent**
Cai et al.(10) **Patent No.:** **US 11,384,105 B2**
(45) **Date of Patent:** ***Jul. 12, 2022**(54) **PROCESSES FOR PREPARING OLIGOMERS**(71) Applicant: **Sarepta Therapeutics, Inc.**,
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Cambridge, MA (US)(*) Notice: Subject to any disclaimer, the term of this
patent is extended or adjusted under 35
U.S.C. 154(b) by 0 days.This patent is subject to a terminal dis-
claimer.(21) Appl. No.: **17/098,935**(22) Filed: **Nov. 16, 2020**(65) **Prior Publication Data**

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Related U.S. Application Data(63) Continuation of application No. 16/303,356, filed as
application No. PCT/US2017/034235 on May 24,
2017, now Pat. No. 10,875,880.(60) Provisional application No. 62/508,256, filed on May
18, 2017, provisional application No. 62/341,049,
filed on May 24, 2016, provisional application No.
62/356,923, filed on Jun. 30, 2016, provisional
application No. 62/357,072, filed on Jun. 30, 2016,
provisional application No. 62/357,153, filed on Jun.
30, 2016, provisional application No. 62/357,166,
filed on Jun. 30, 2016.(51) **Int. Cl.**
C07F 9/6561 (2006.01)
C07F 9/6558 (2006.01)(52) **U.S. Cl.**
CPC **C07F 9/65616** (2013.01); **C07F 9/65583**
(2013.01)(58) **Field of Classification Search**
CPC C07F 9/65616; C07F 9/65583
See application file for complete search history.(56) **References Cited**

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Primary Examiner — Kamal A Saeed(74) *Attorney, Agent, or Firm* — Sterne, Kessler,
Goldstein Fox(57) **ABSTRACT**Provided herein are processes for preparing an oligomer
(e.g., a morpholino oligomer). The synthetic processes
described herein may be advantageous to scaling up oli-
gomer synthesis while maintaining overall yield and purity
of a synthesized oligomer.**8 Claims, 3 Drawing Sheets****Specification includes a Sequence Listing.**

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Fig. 1

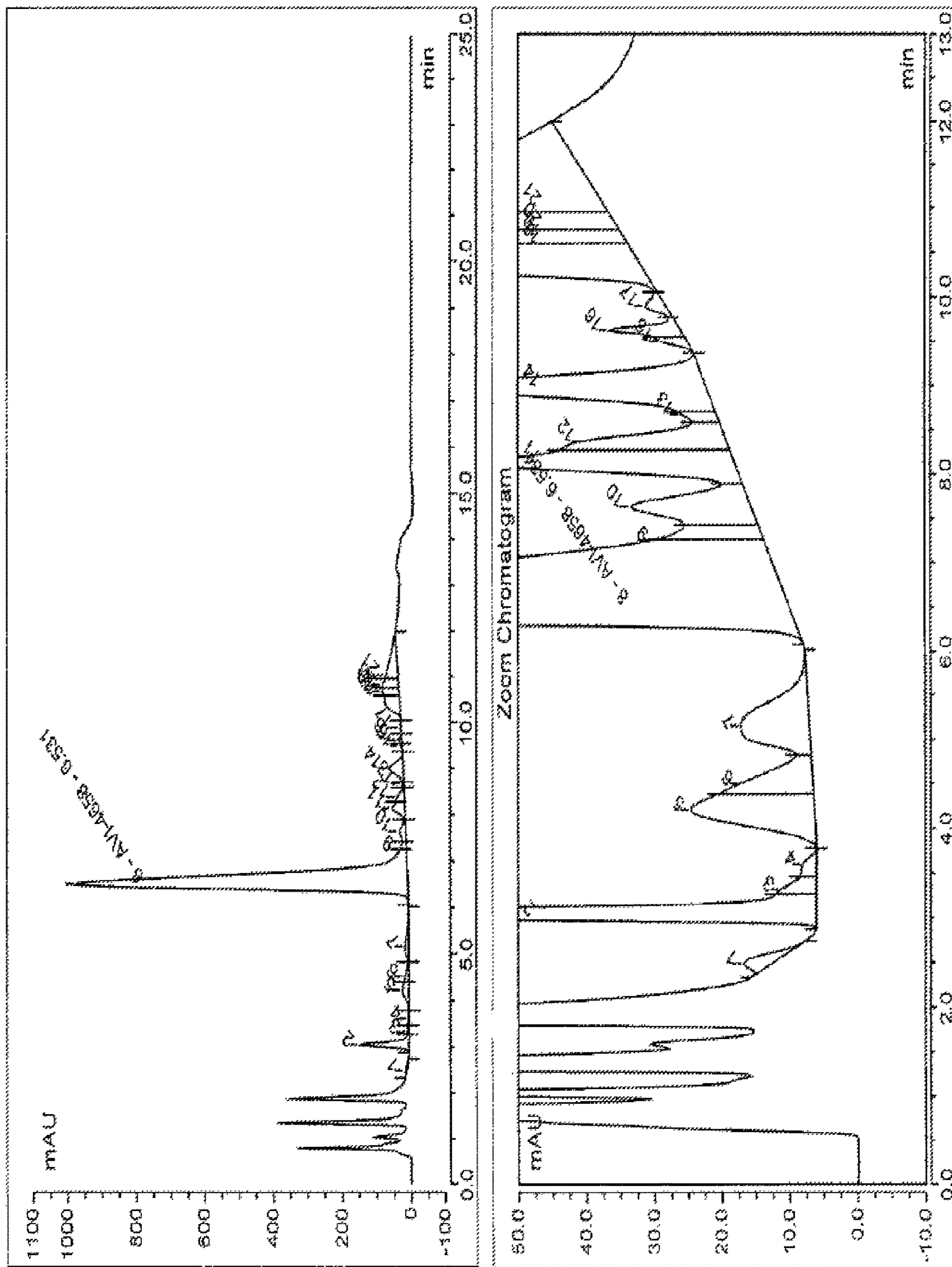


Fig. 2

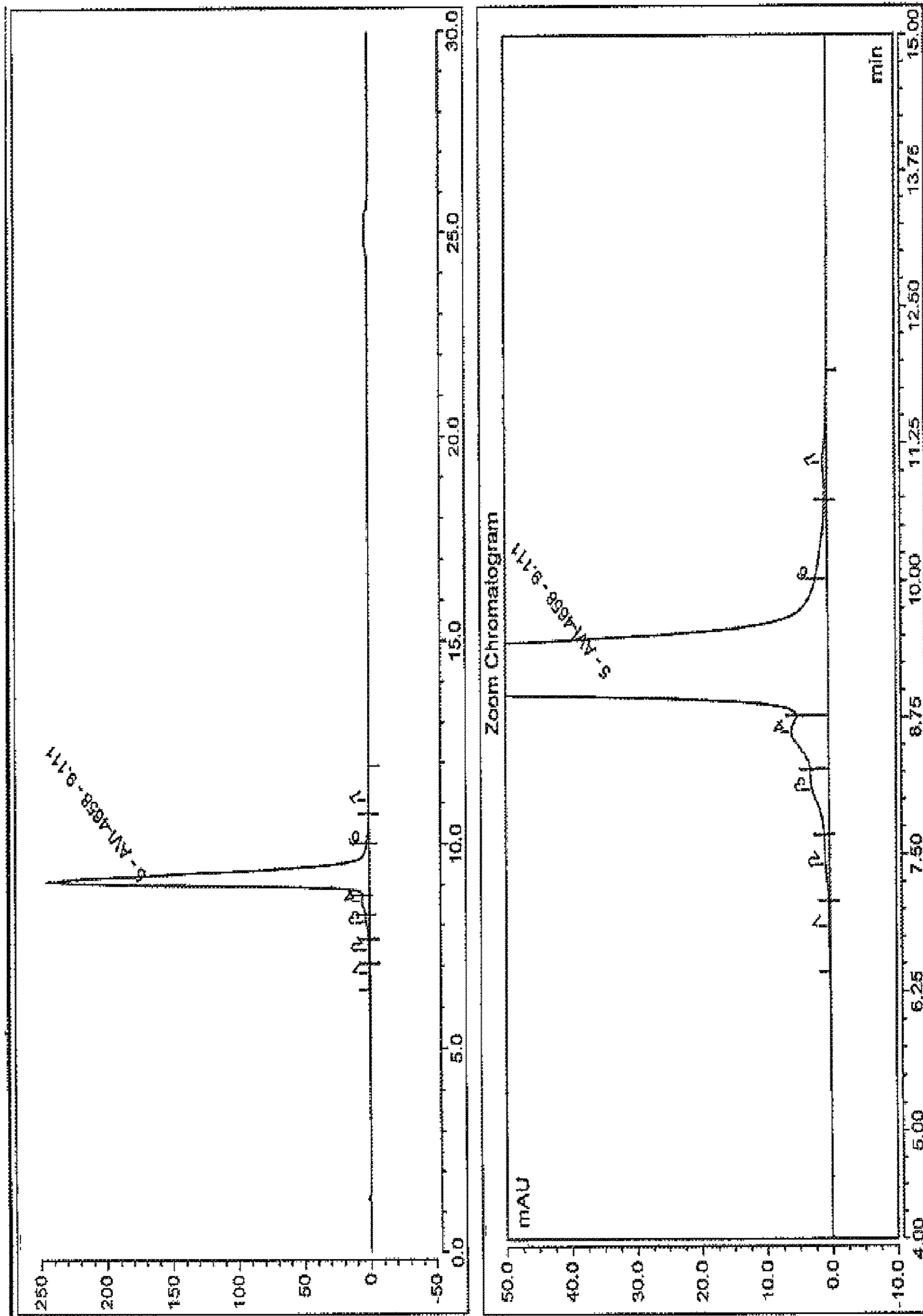
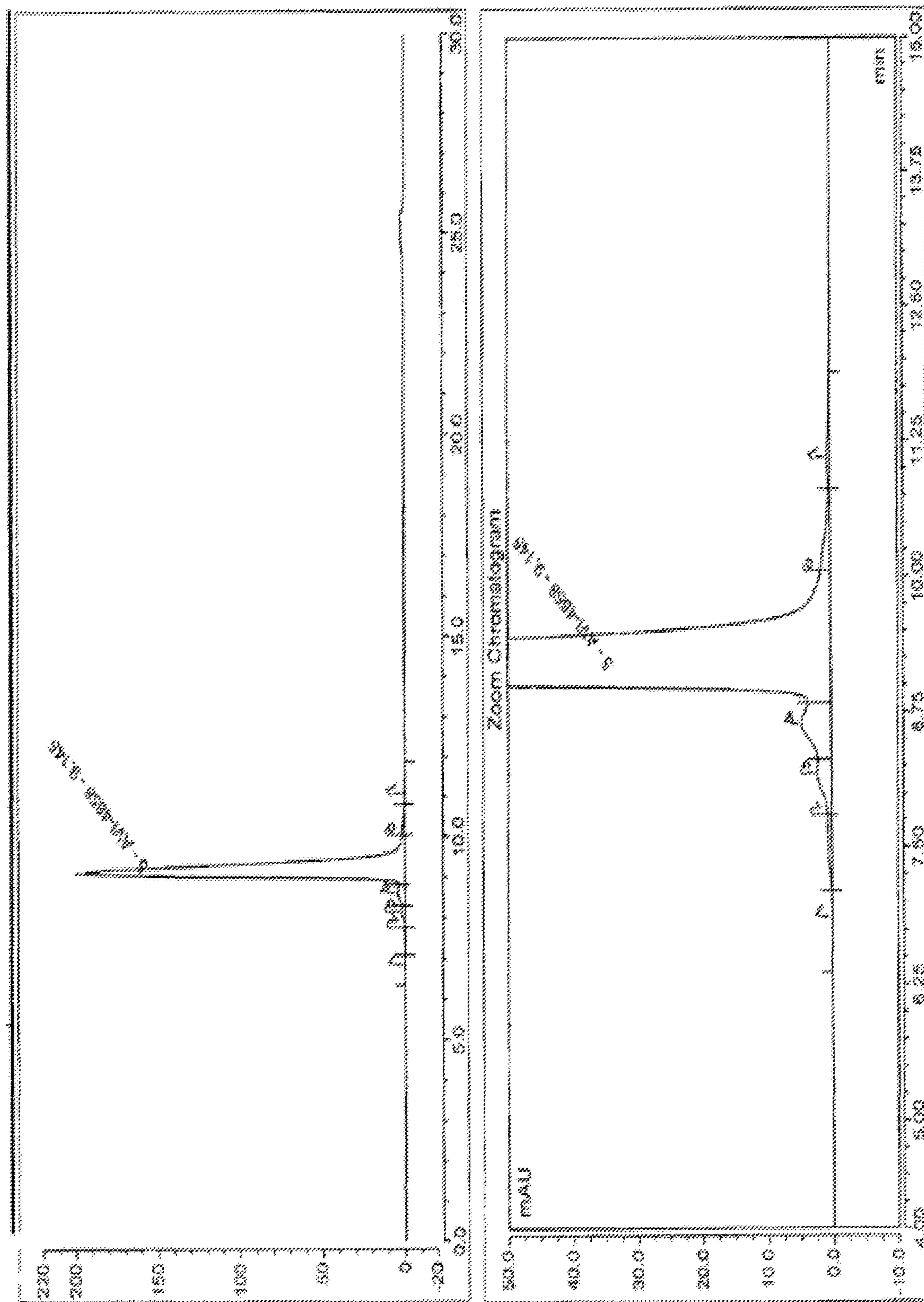


Fig. 3



PROCESSES FOR PREPARING OLIGOMERS

RELATED APPLICATIONS

This application claims benefit of U.S. provisional patent application No. 62/508,256, filed May 18, 2017; U.S. provisional patent application No. 62/341,049 filed May 24, 2016; U.S. provisional patent application No. 62/356,923, filed Jun. 30, 2016; U.S. provisional patent application No. 62/357,072, filed Jun. 30, 2016; U.S. provisional patent application No. 62/357,153, filed Jun. 30, 2016; and U.S. provisional patent application No. 62/357,166, filed Jun. 30, 2016; the entire contents of each of which are incorporated herein by reference.

REFERENCE TO SEQUENCE LISTING
SUBMITTED ELECTRONICALLY

The content of the electronically submitted sequence listing (Name: 4140_0360007_Seglisting_ST25; Size: 2,556 bytes; and Date of Creation: Mar. 5, 2021) is herein incorporated by reference in its entirety.

BACKGROUND

Antisense technology provides a means for modulating the expression of one or more specific gene products, including alternative splice products, and is uniquely useful in a number of therapeutic, diagnostic, and research applications. The principle behind antisense technology is that an antisense compound, e.g., an oligonucleotide, which hybridizes to a target nucleic acid, modulates gene expression activities such as transcription, splicing or translation through any one of a number of antisense mechanisms. The sequence specificity of antisense compounds makes them attractive as tools for target validation and gene functionalization, as well as therapeutics to selectively modulate the expression of genes involved in disease.

Duchenne muscular dystrophy (DMD) is caused by a defect in the expression of the protein dystrophin. The gene encoding the protein contains 79 exons spread out over more than 2 million nucleotides of DNA. Any exonic mutation that changes the reading frame of the exon, or introduces a stop codon, or is characterized by removal of an entire out of frame exon or exons, or duplications of one or more exons, has the potential to disrupt production of functional dystrophin, resulting in DMD.

Recent clinical trials testing the safety and efficacy of splice switching oligonucleotides (SSOs) for the treatment of DMD are based on SSO technology to induce alternative splicing of pre-mRNAs by steric blockade of the spliceosome (Cirak et al., 2011; Goemans et al., 2011; Kinali et al., 2009; van Deutekom et al., 2007). However, despite these successes, the pharmacological options available for treating DMD are limited.

Eteplirsen is a phosphorodiamidate morpholino oligomer (PMO) designed to skip exon 51 of the human dystrophin gene in patients with DMD who are amendable to exon 51

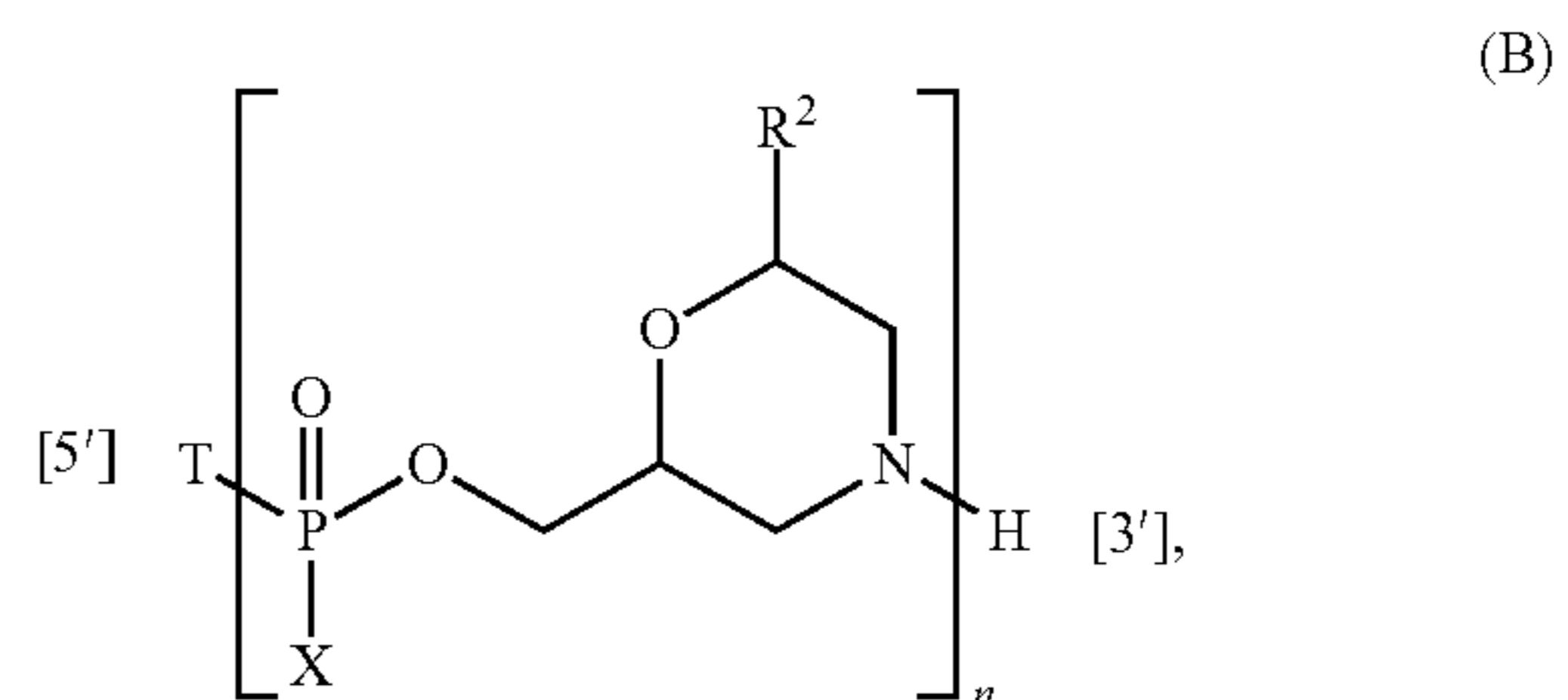
skipping to restore the read frame and produce a functional shorter form of the dystrophin protein. Sarepta Therapeutics, Inc., submitted a New Drug Application (NDA) to the United States Food and Drug Administration (FDA) seeking approval for the treatment of DMD in patients amendable to exon 51 skipping. Sarepta's NDA is currently under review by the FDA.

Although significant progress has been made in the field of antisense technology, there remains a need in the art for methods of preparing phosphorodiamidate morpholino oligomers with improved antisense or antigene performance.

SUMMARY

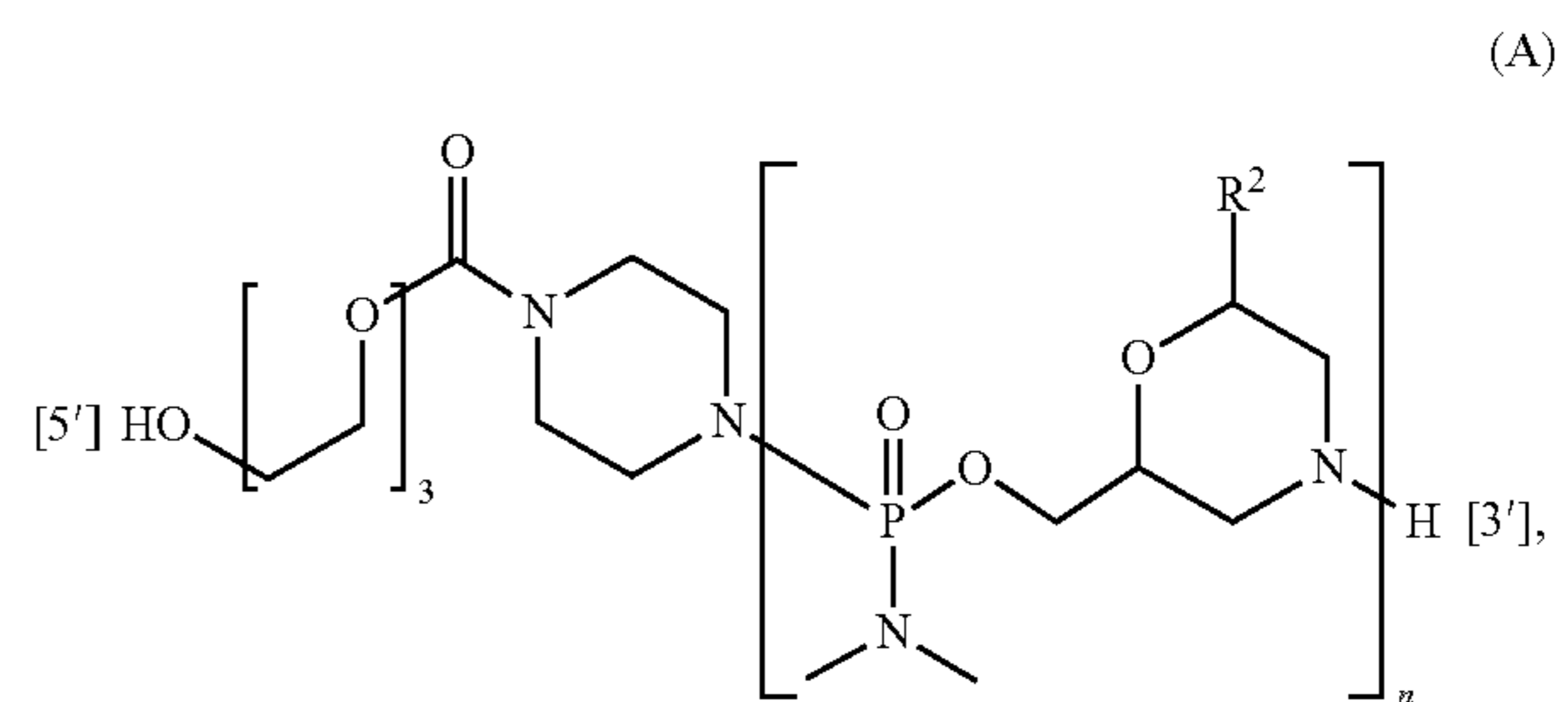
Provided herein are processes for preparing phosphorodiamidate morpholino oligomers (PMOs). The synthetic processes described herein allow for a scaled-up PMO synthesis while maintaining overall yield and purity of a synthesized PMO.

In one aspect, the disclosure provides a process for preparing an oligomeric compound of Formula (B):



or a pharmaceutically acceptable salt thereof, wherein T, X, R², and n are as defined herein.

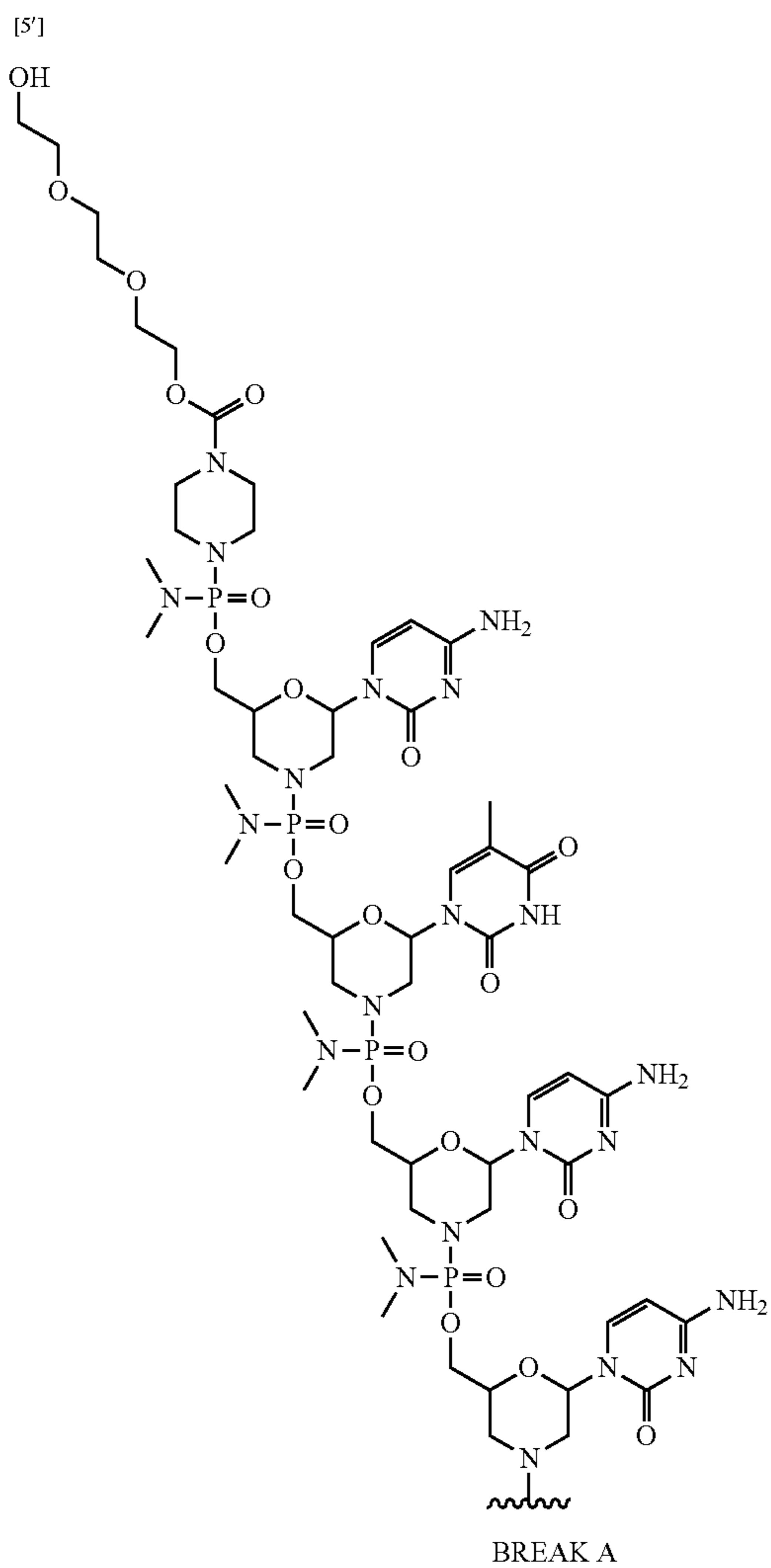
In some embodiments, the disclosure provides a process for preparing an oligomeric compound of Formula (A):



or a pharmaceutically acceptable salt thereof, wherein R² and n are as defined herein.

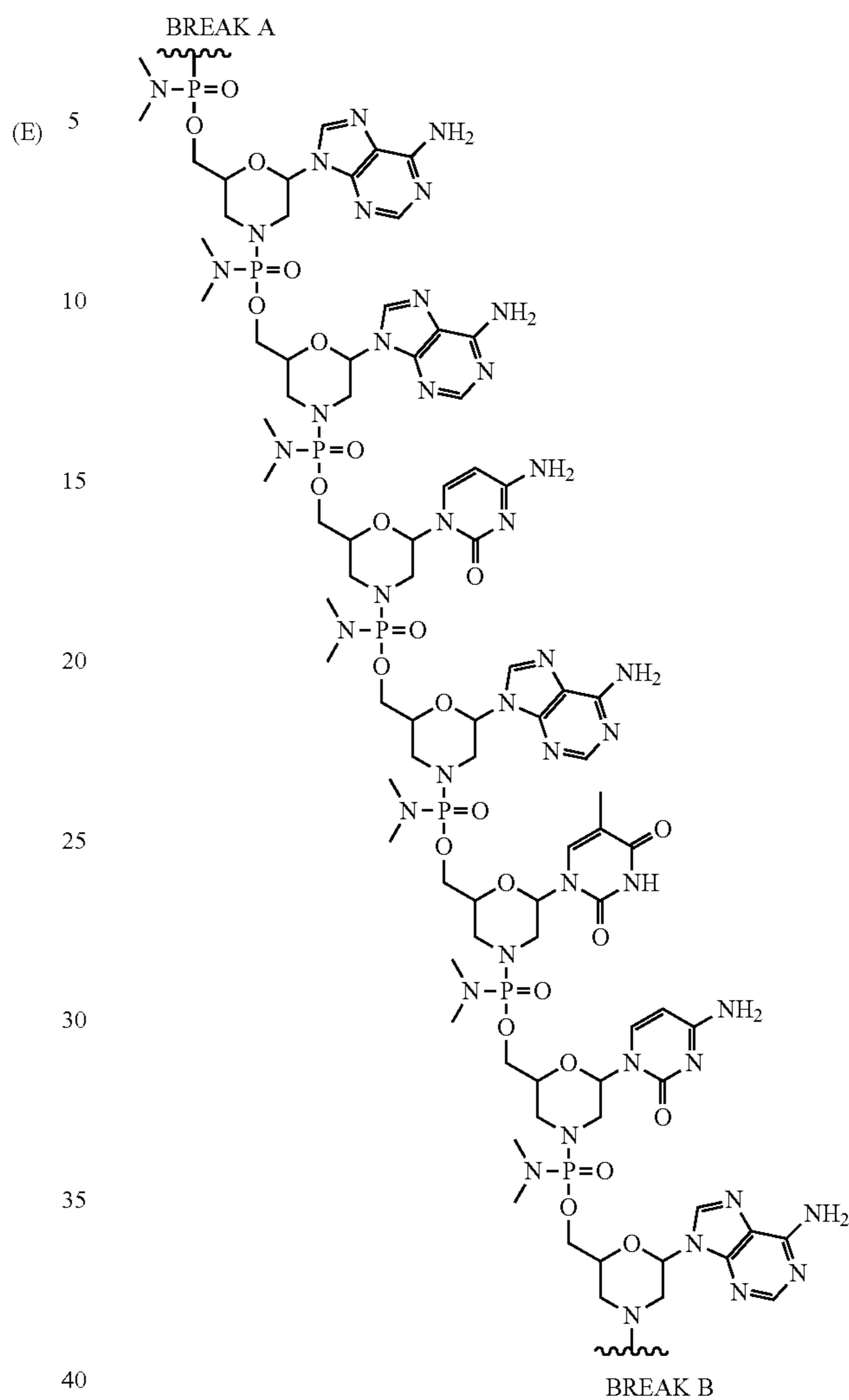
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In certain embodiments, provided herein is a process for preparing an oligomeric compound of Formula (E):



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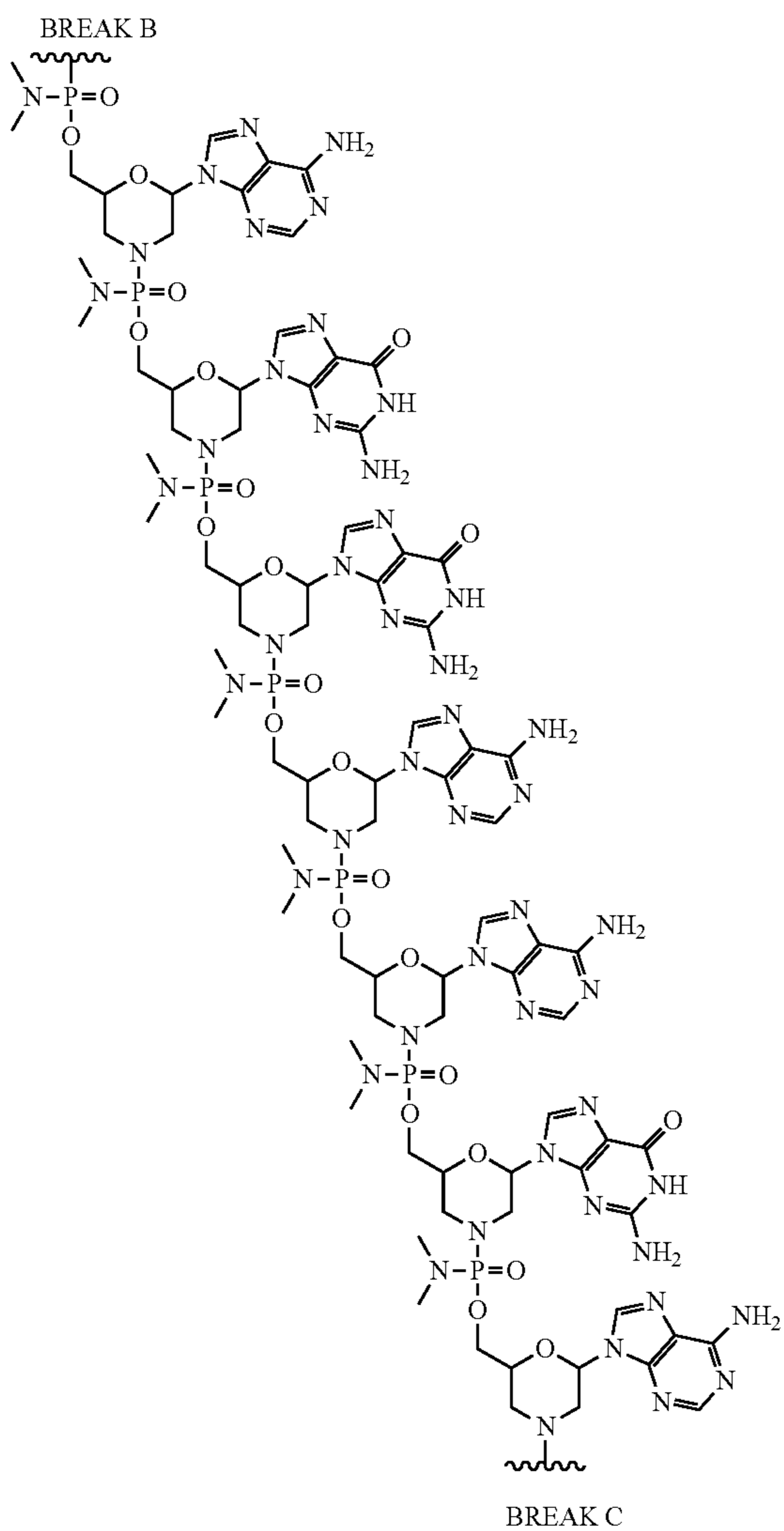
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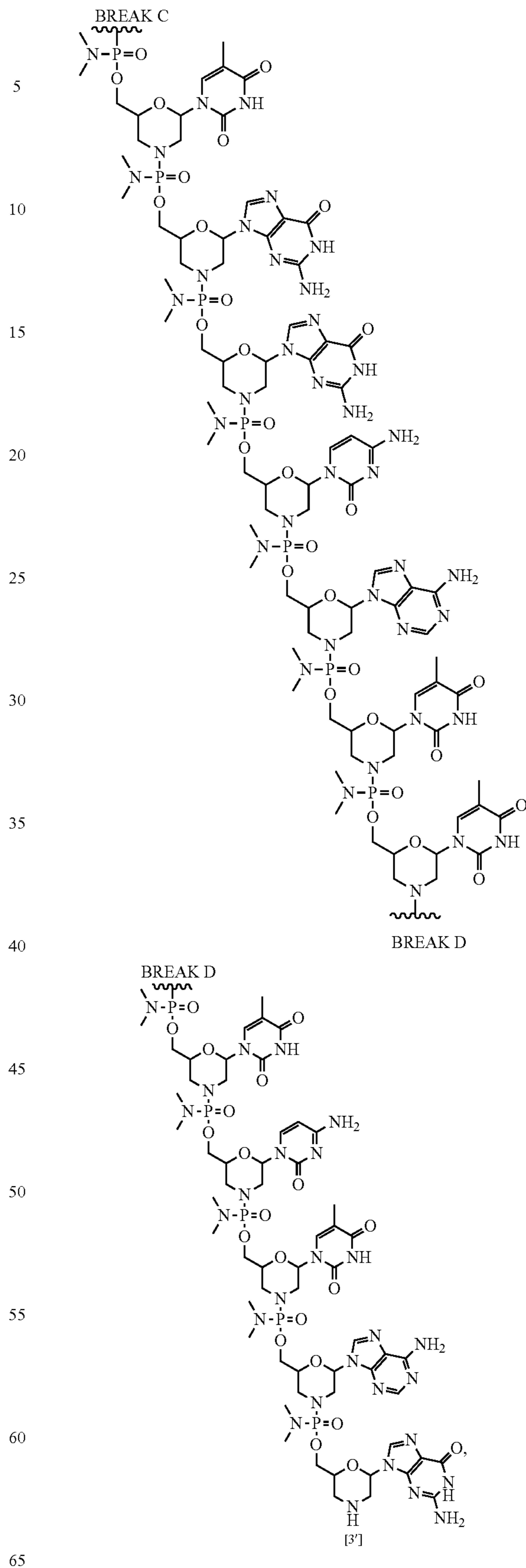
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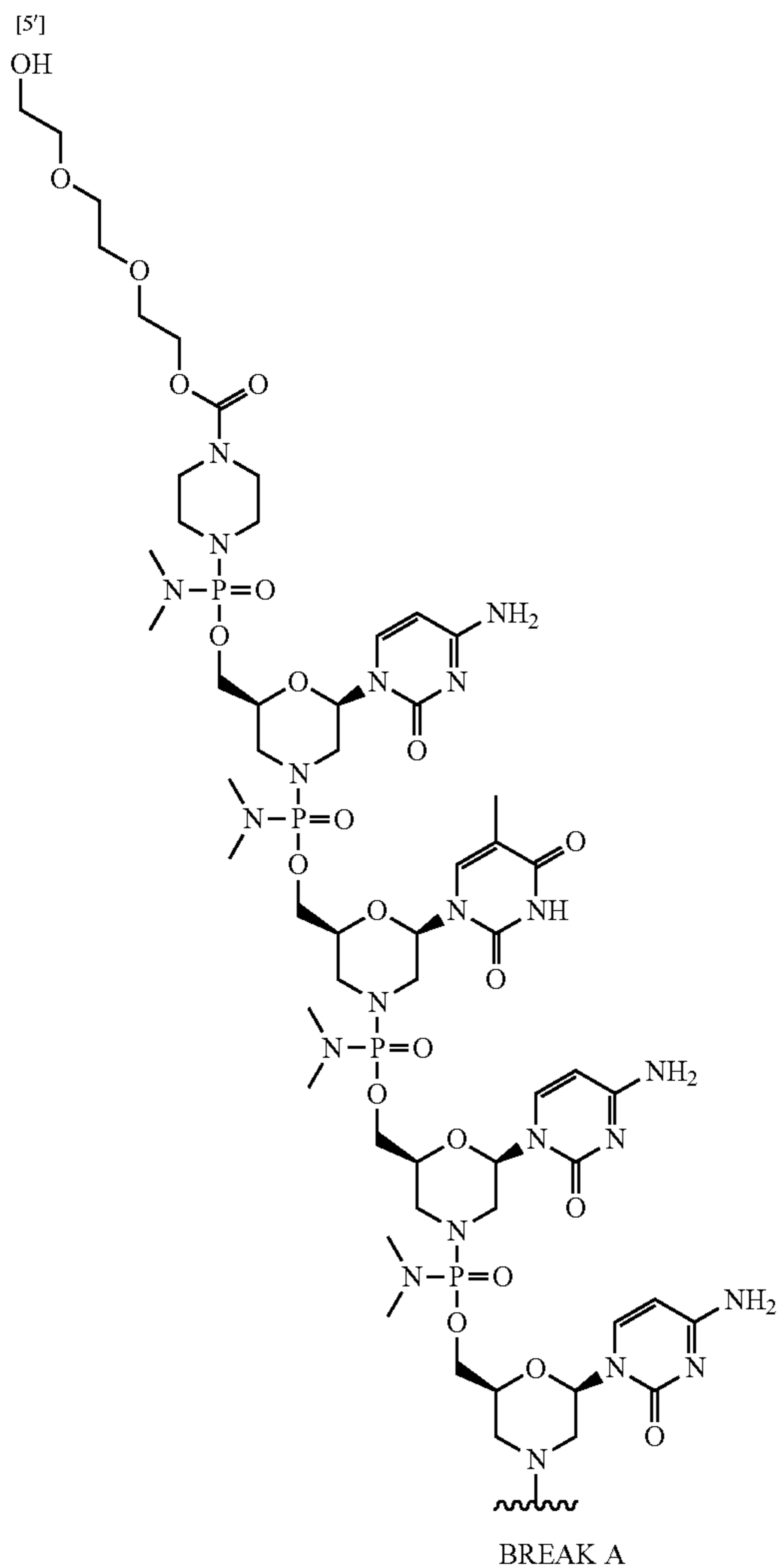
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or a pharmaceutically acceptable salt thereof.

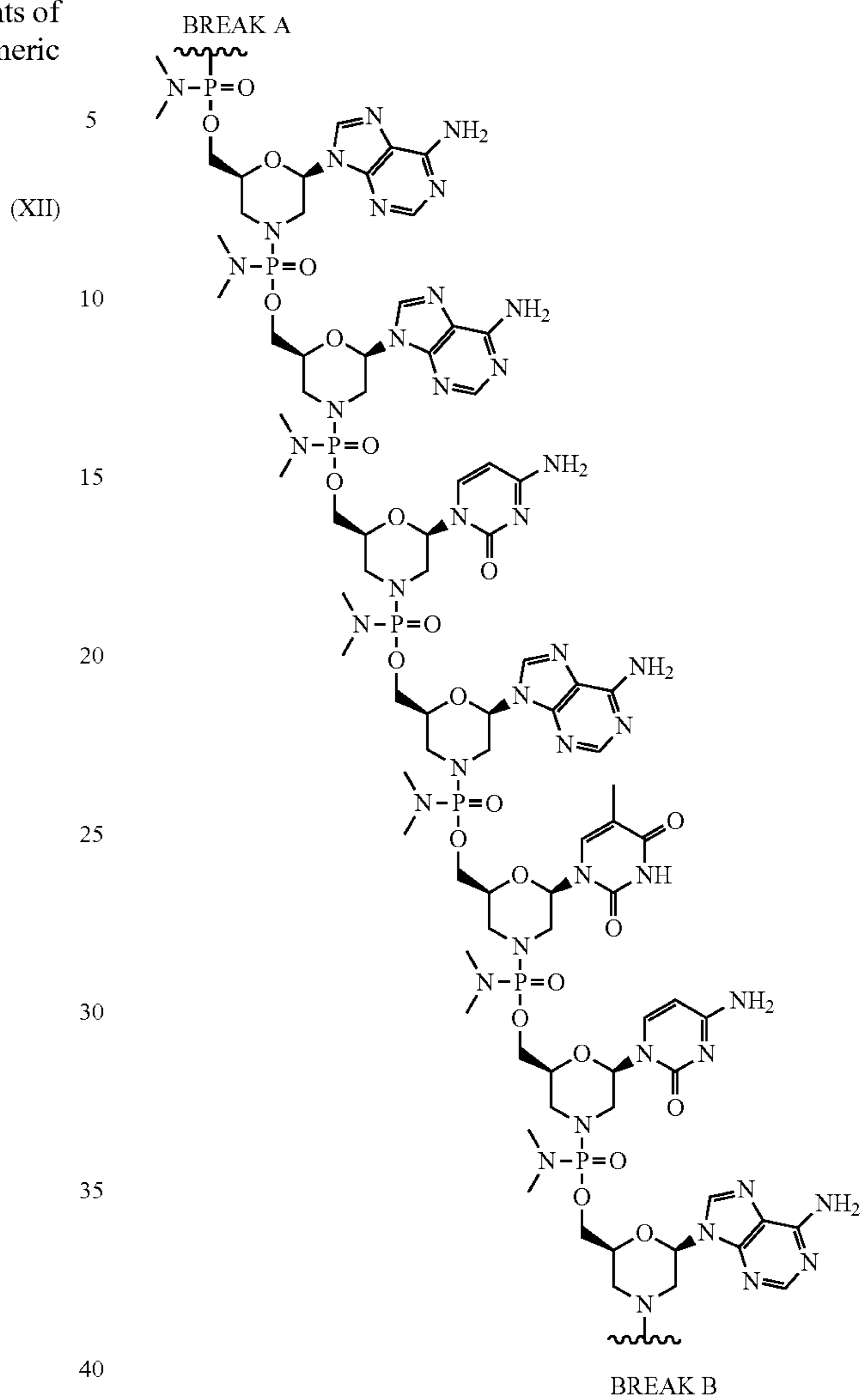
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In yet another embodiment, the oligomeric compound of the disclosure including, for example, some embodiments of an oligomeric compound of Formula (E), is an oligomeric compound of Formula (XII):



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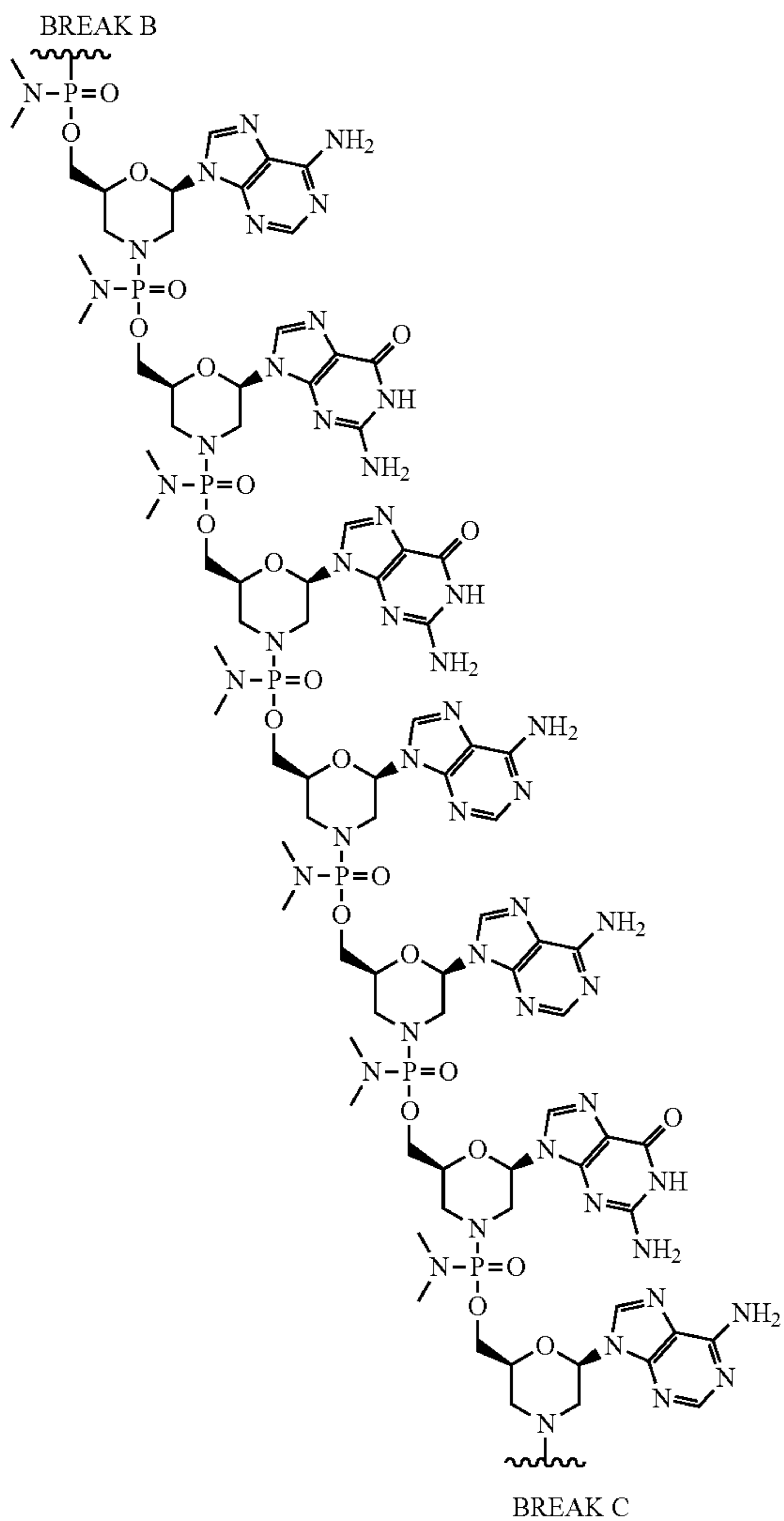
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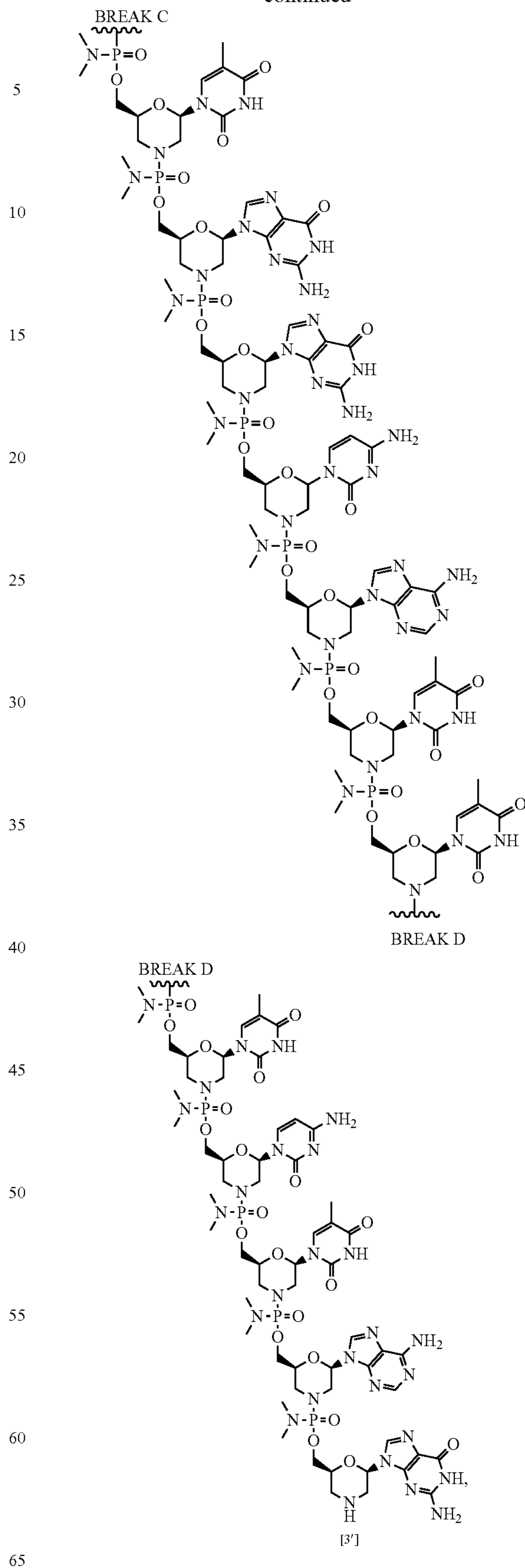
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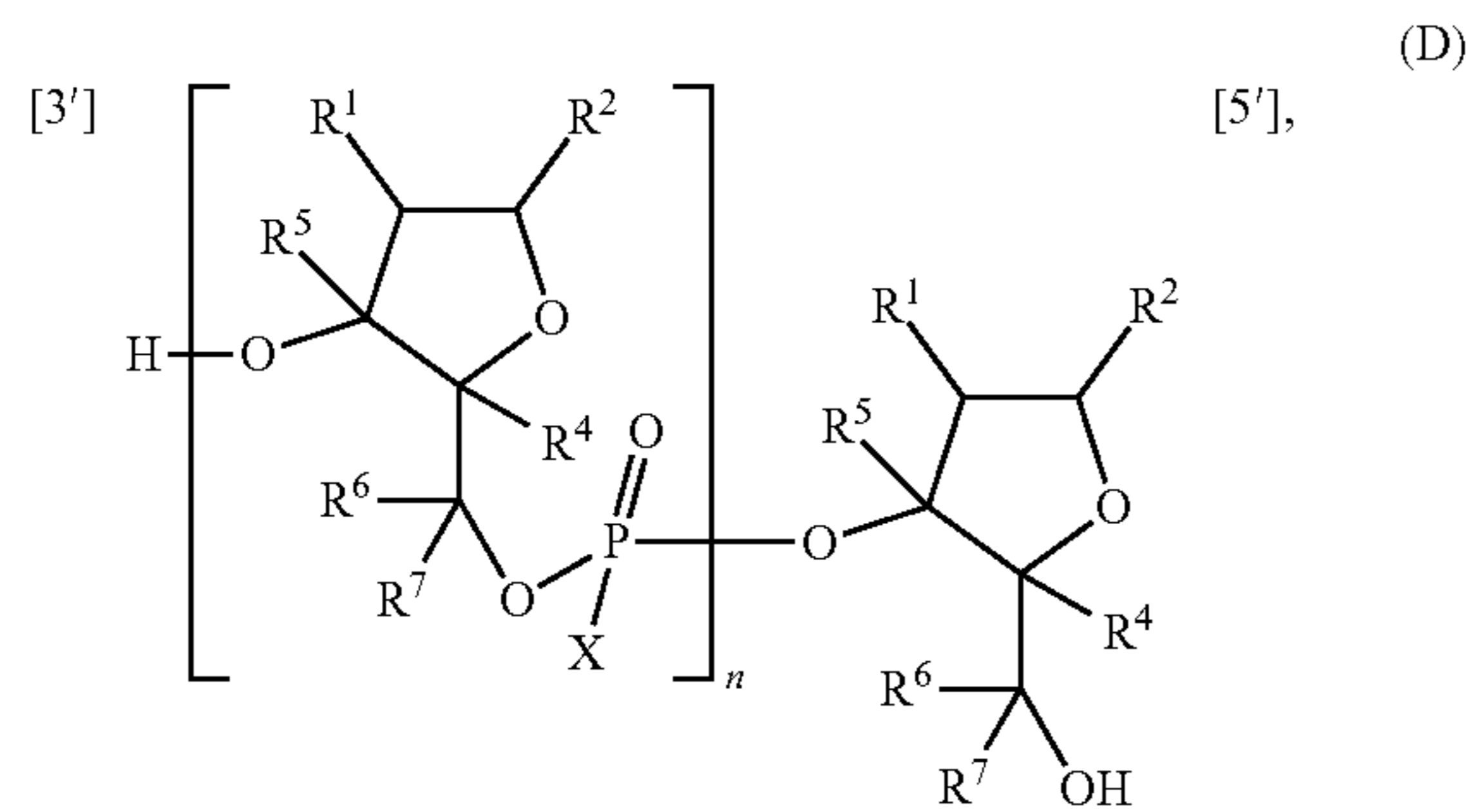


or a pharmaceutically acceptable salt thereof.

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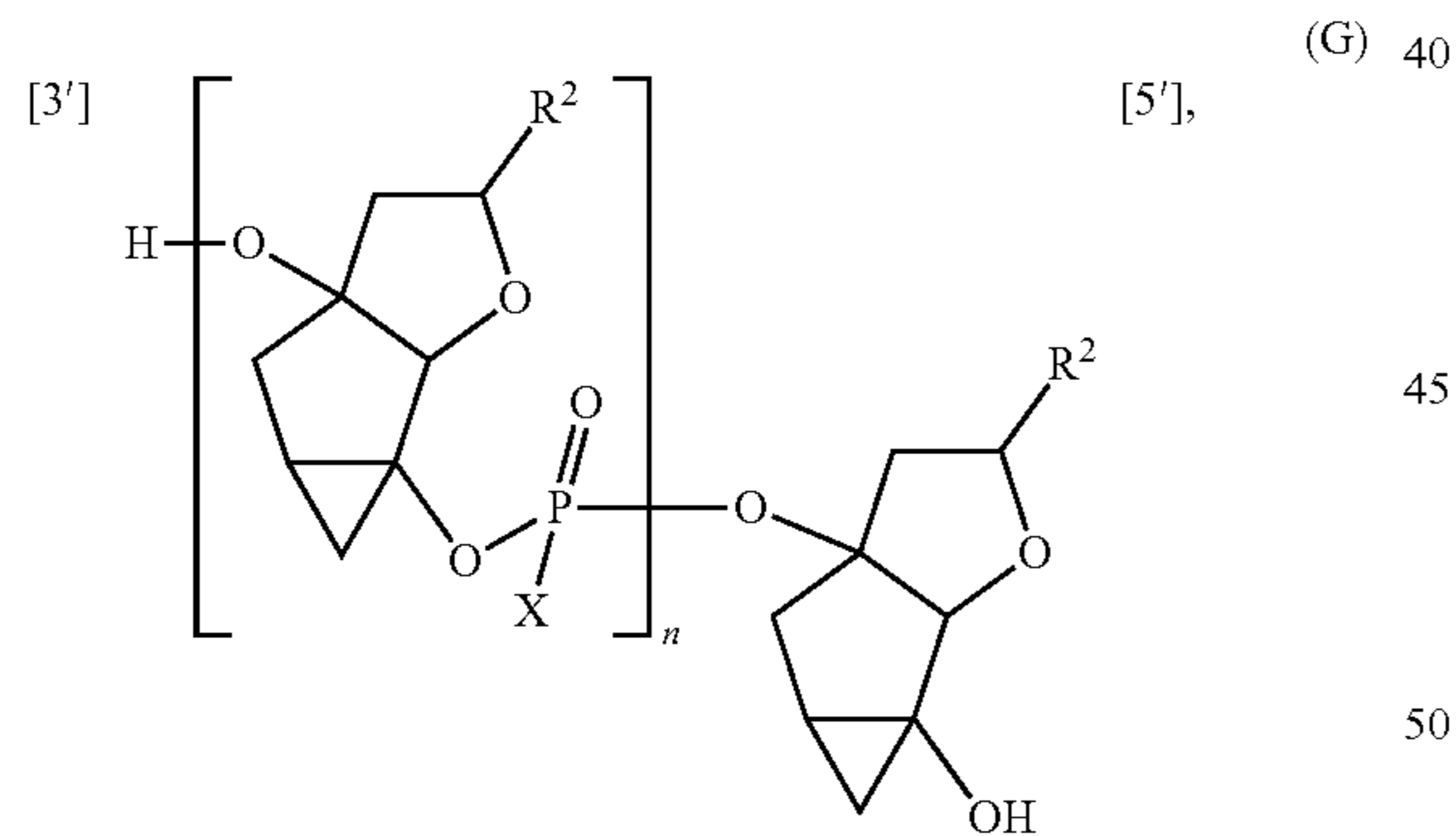
For clarity, the structural formulas including, for example, oligomeric compound of Formula (E) and Eteplirsen depicted by Formula (XII), are a continuous structural formula from 5' to 3', and, for the convenience of depicting the entire formula in compact form in the above structural formulas, Applicants have included various illustration breaks labeled "BREAK A," "BREAK B," "BREAK C," and "BREAK D." As would be understood by the skilled artisan, for example, each indication of "BREAK A" shows a continuation of the illustration of the structural formula at these points. The skilled artisan understands that the same is true for each instance of "BREAK B," "BREAK C," and "BREAK D" in the structural formulas above including Eteplirsen. None of the illustration breaks, however, are intended to indicate, nor would the skilled artisan understand them to mean, an actual discontinuation of the structural formulas above including Eteplirsen.

In another aspect of the disclosure is provided a process for preparing an oligomeric compound of Formula (D):



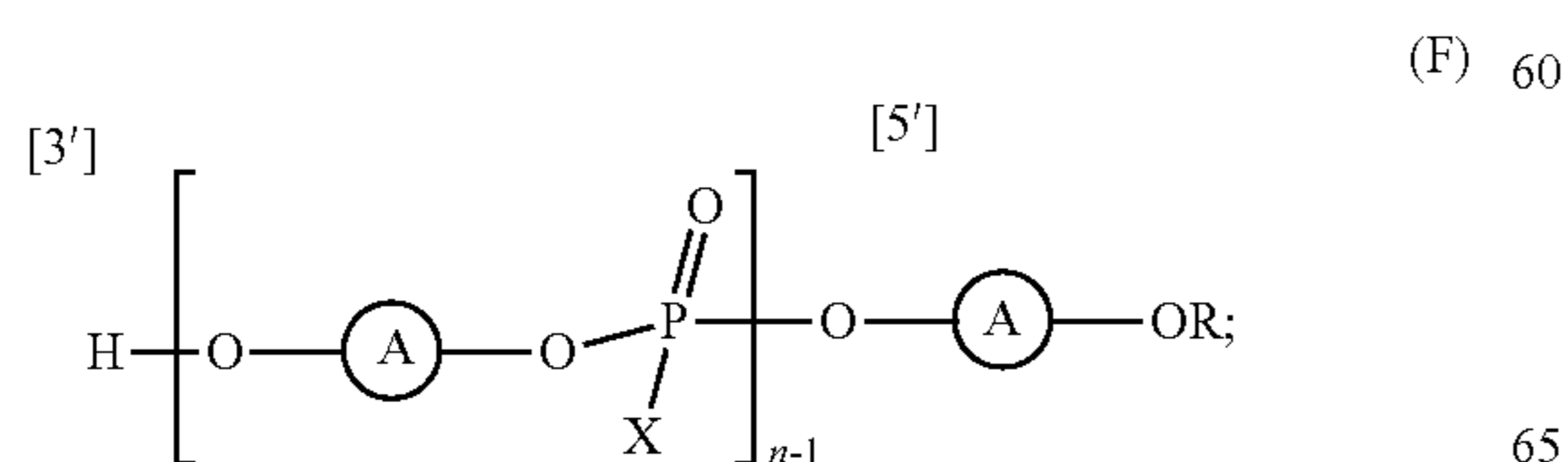
or a pharmaceutically acceptable salt thereof, wherein R¹, R², R⁴-R⁷, X, and n are as defined herein.

In some embodiments, the oligomeric compound of Formula (D) is of Formula (G):



or a pharmaceutically acceptable salt thereof, wherein R², X, and n are as defined herein.

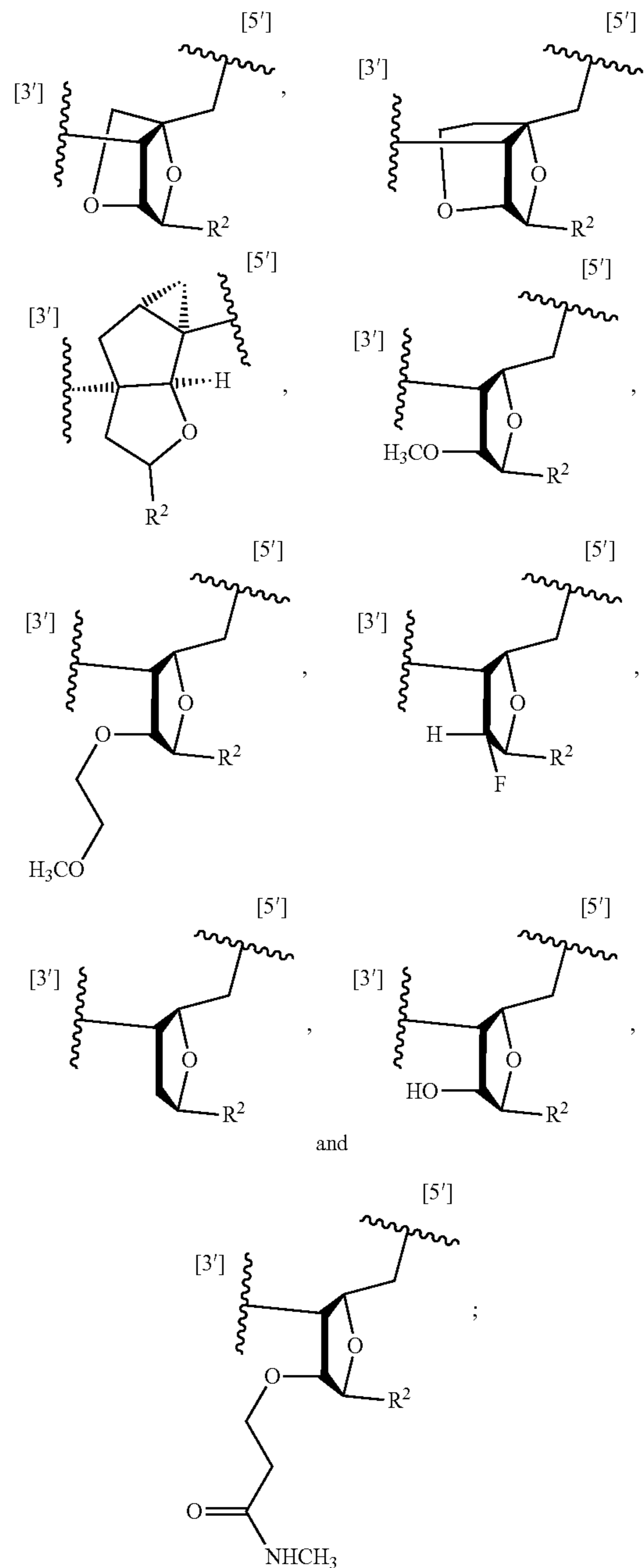
In another aspect of the disclosure is provided a process for preparing an oligomeric compound of Formula (F):



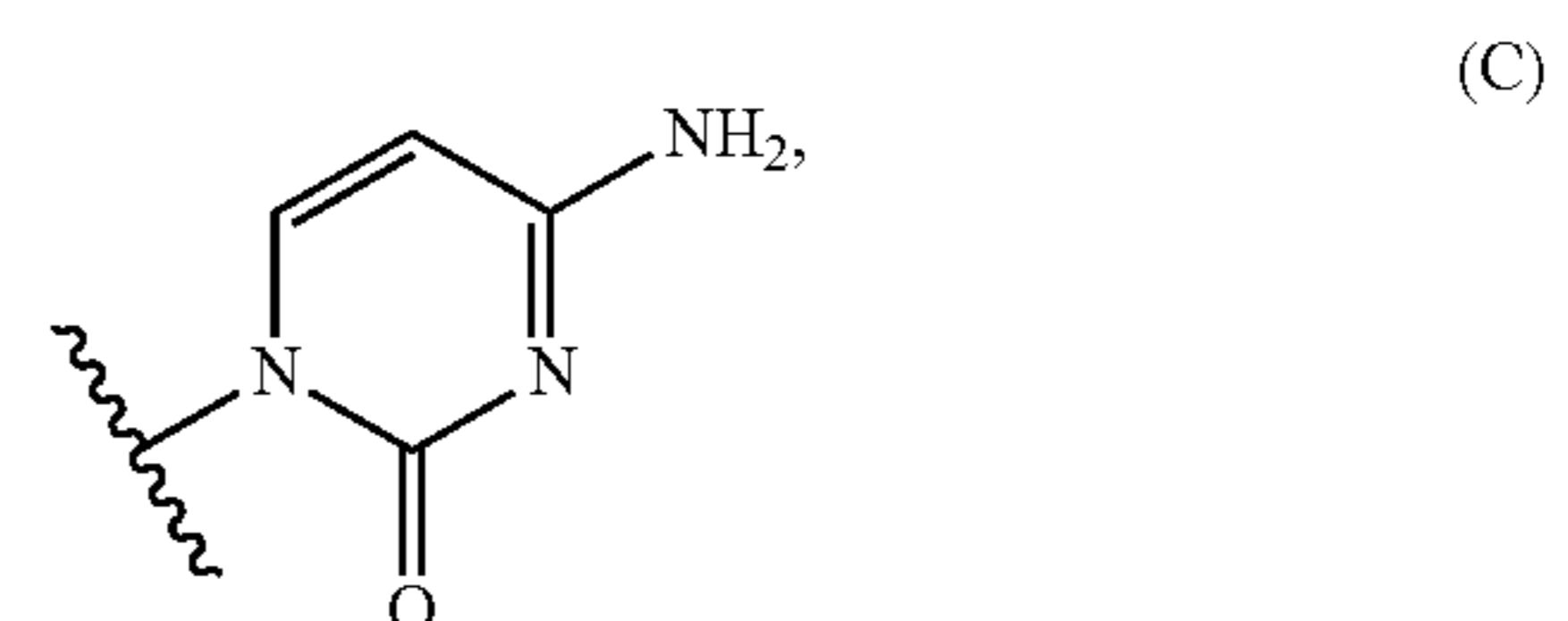
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wherein:

each A is, independently for each occurrence, selected from the group consisting of:

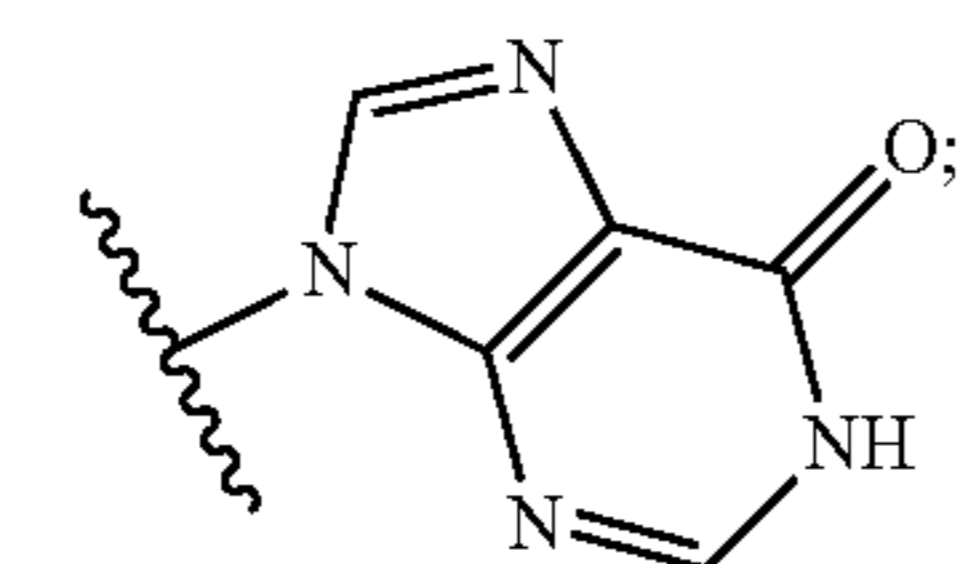
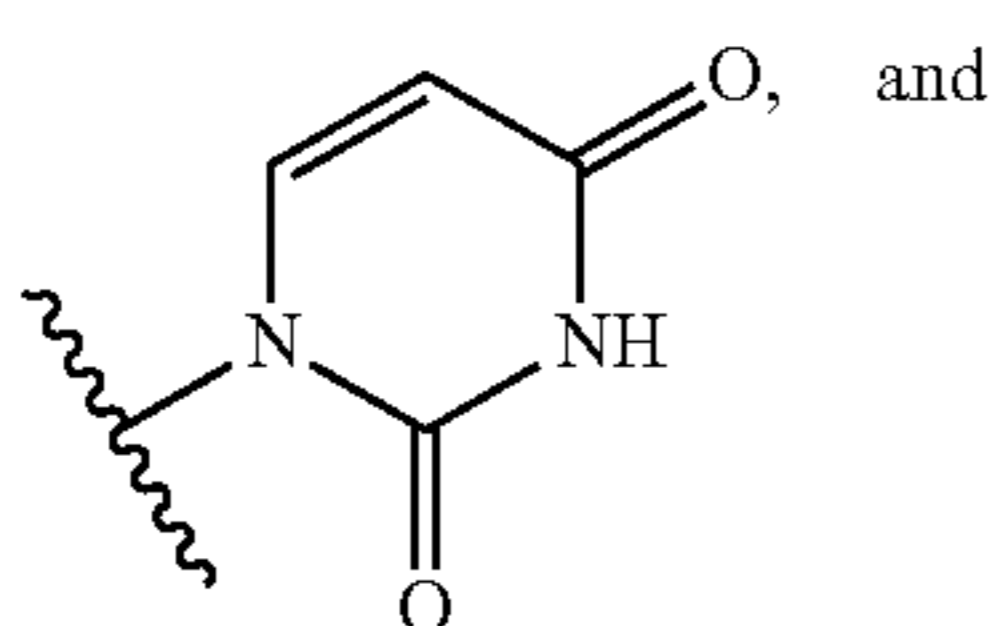
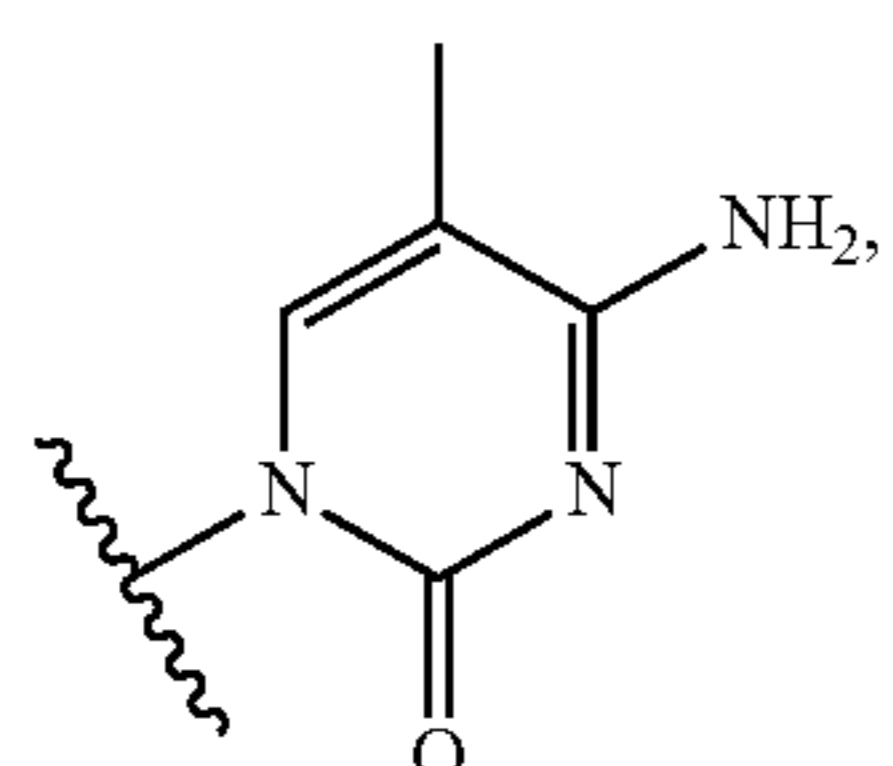
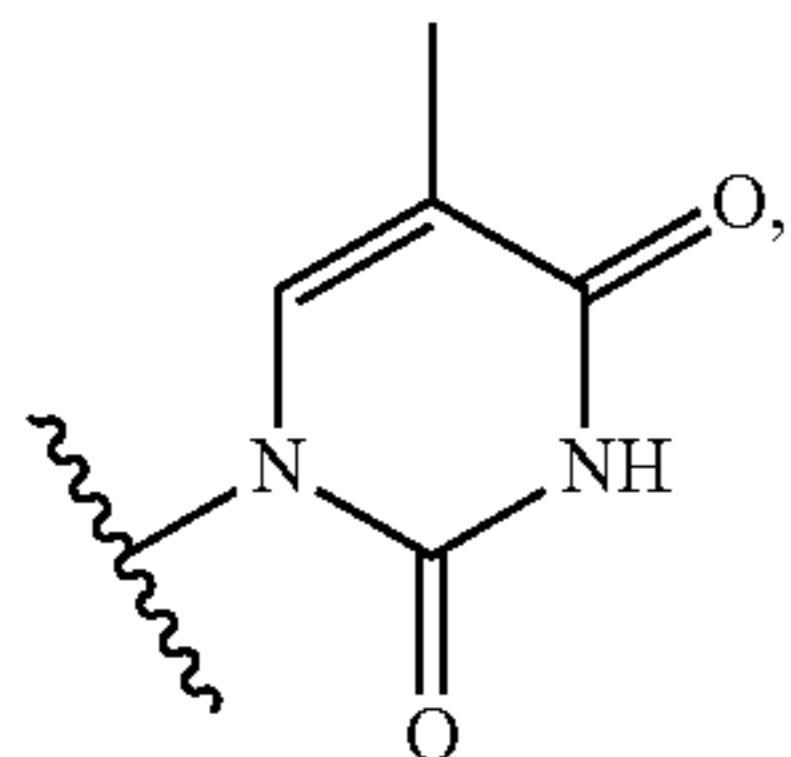
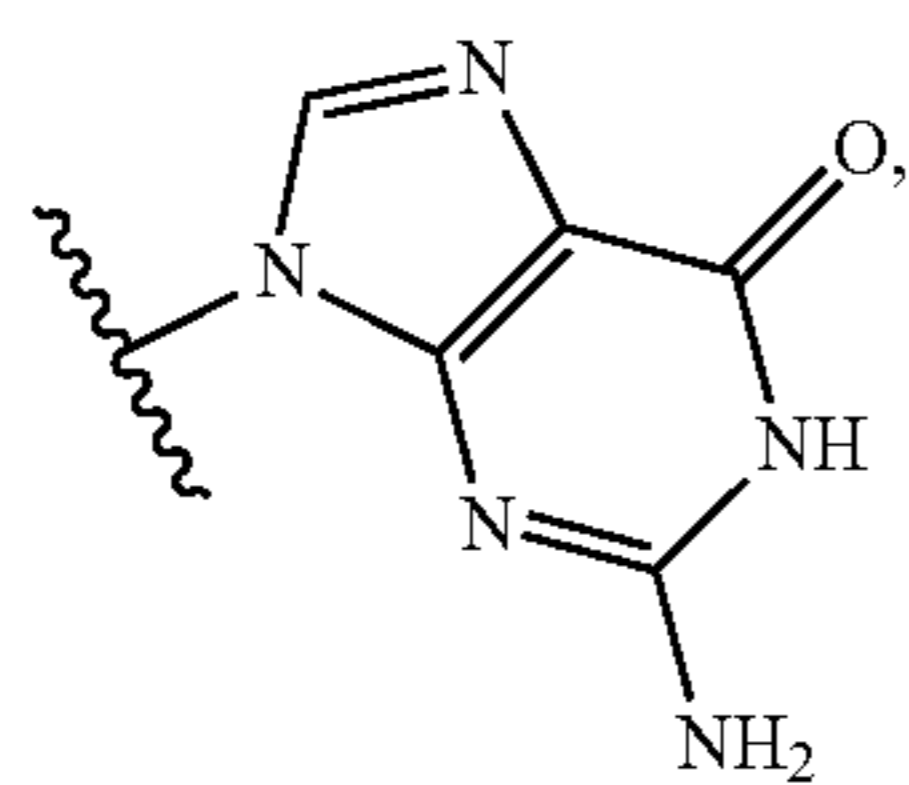


each R² is, independently for each occurrence, selected from the group consisting of:



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-continued



n is an integer from 10 to 40; and
R is H or acetyl.

BRIEF DESCRIPTION OF THE FIGURES

FIG. 1 shows a representative analytical high performance liquid chromatography (HPLC) chromatogram of a synthesized and deprotected Eteplirsen (AVI-4658) crude drug substance (see Example 4).

FIG. 2 shows a representative analytical HPLC chromatogram of a purified Eteplirsen drug substance solution (see Example 5).

FIG. 3 shows a representative analytical HPLC chromatogram of a desalted and lyophilized Eteplirsen drug substance (see Example 5).

DETAILED DESCRIPTION

Provided herein are processes for preparing a morpholino oligomer. The a morpholino oligomer described herein displays stronger affinity for DNA and RNA without compromising sequence selectivity, relative to native or unmodified

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oligonucleotides. In some embodiments, the morpholino oligomer of the disclosure minimizes or prevents cleavage by RNase H. In some embodiments, the morpholino oligomer of the disclosure does not activate RNase H.

The processes described herein are advantageous in an industrial-scale process and can be applied to preparing quantities of a morpholino oligomer in high yield and scale (e.g., about 1 kg, about 1-10 kg, about 2-10 kg, about 5-20 kg, about 10-20 kg, or about 10-50 kg).

Definitions

Listed below are definitions of various terms used to describe this disclosure. These definitions apply to the terms as they are used throughout this specification and claims, unless otherwise limited in specific instances, either individually or as part of a larger group.

“Base-protected” or “base protection” refers to protection of the base-pairing groups, e.g., purine or pyrimidine bases, on the morpholino subunits with protecting groups suitable to prevent reaction or interference of the base-pairing groups during stepwise oligomer synthesis. An example of a base-protected morpholino subunit is the activated C subunit Compound (C) having a CBZ protecting group on the cytosine amino group depicted below.

An “activated phosphoramidate group” is typically a chlorophosphoramidate group, having substitution at nitrogen which is desired in the eventual phosphordiamidate linkage in the oligomer. An example is (dimethylamino) chlorophosphoramidate, i.e., $-\text{O}-\text{P}(=\text{O})(\text{NMe}_2)\text{Cl}$.

The term “support-bound” refers to a chemical entity that is covalently linked to a support-medium.

The term “support-medium” refers to any material including, for example, any particle, bead, or surface, upon which an oligomer can be attached or synthesized upon, or can be modified for attachment or synthesis of an oligomer. Representative substrates include, but are not limited to, inorganic supports and organic supports such as glass and modified or functionalized glass, plastics (including acrylics, polystyrene and copolymers of styrene and other materials, polypropylene, polyethylene, polybutylene, polyurethanes, TEFLON, etc.), polysaccharides, nylon or nitrocellulose, ceramics, resins, silica or silica-based materials including silicon and modified silicon, carbon, metals, inorganic glasses, plastics, optical fiber bundles, and a variety of other polymers. Particularly useful support-medium and solid surfaces for some embodiments are located within a flow cell apparatus. In some embodiments of the processes described herein, the support-medium comprises polystyrene with 1% crosslinked divinylbenzene.

In some embodiments, representative support-medium comprise at least one reactive site for attachment or synthesis of an oligomer. For example, in some embodiments, a support-medium of the disclosure comprises one or more terminal amino or hydroxyl groups capable of forming a chemical bond with an incoming subunit or other activated group for attaching or synthesizing an oligomer.

Some representative support-media that are amenable to the processes described herein include, but are not limited to, the following: controlled pore glass (CPG); oxalyl-controlled pore glass (see, e.g., Alul et al., *Nucleic Acids Research* 1991, 19, 1527); silica-containing particles, such as porous glass beads and silica gel such as that formed by the reaction of trichloro-[3-(4-chloromethyl)phenyl]propylsilane and porous glass beads (see Parr and Grohmann, *Angew. Chem. Internat. Ed.* 1972, 11, 314, sold under the trademark “PORASIL E” by Waters Associates, Framing-

ham, Mass., USA); a mono ester of 1,4-dihydroxymethylbenzene and silica (see Bayer and Jung, *Tetrahedron Lett.* 1970, 51, 4503, sold under the trademark "BIOPAK" by Waters Associates); TENTAGEL (see, e.g., Wright et al., *Tetrahedron Lett.* 1993, 34, 3373); cross-linked styrene/divinylbenzene copolymer beaded matrix, or POROS, a copolymer of polystyrene/divinylbenzene (available from PerSeptive Biosystems); soluble support-medium such as polyethylene glycol PEG's (see Bonora et al., *Organic Process Research & Development* 2000, 4, 225-231); PEPS support, which is a polyethylene (PE) film with pendant long-chain polystyrene (PS) grafts (see Berg et al., *J. Am. Chem. Soc.* 1989, 111, 8024 and International Patent Application WO 1990/02749); copolymers of dimethylacrylamide cross-linked with N,N'-bisacryloyl ethylenediamine, including a known amount of N-tertbutoxycarbonyl-beta-alanyl-N'-acryloylhexamethylenediamine (see Atherton et al., *J. Am. Chem. Soc.* 1975, 97, 6584, Atherton et al., *Bioorg. Chem.* 1979, 8, 351, and Atherton et al., *J. Chem. Soc. Perkin I* 1981, 538); glass particles coated with a hydrophobic cross-linked styrene polymer (see Scott et al., *J. Chrom. Sci.* 1971, 9, 577); fluorinated ethylene polymer onto which has been grafted polystyrene (see Kent and Merrifield, *Israel J. Chem.* 1978, 17, 243 and van Rietschoten in *Peptides* 1974, Y. Wolman, Ed., Wiley and Sons, New York, 1975, pp. 113-116); hydroxypropylacrylate-coated polypropylene membranes (Daniels et al., *Tetrahedron Lett.* 1989, 4345); acrylic acid-grafted polyethylene-rods (Geysen et al., *Proc. Nat. Acad. Sci. USA* 1984, 81, 3998); a "tea bag" containing traditionally-used polymer beads (Houghten, *Proc. Nat. Acad. Sci. USA* 1985, 82, 5131); and combinations thereof.

The term "flow cell apparatus" refers to a chamber comprising a surface (e.g., solid surface) across which one or more fluid reagents (e.g., liquid or gas) can be flowed.

The term "deblocking agent" refers to a composition (e.g., a solution) comprising a chemical acid or combination of chemical acids for removing protecting groups. Exemplary chemical acids used in deblocking agents include halogenated acids, e.g., chloroacetic acid, dichloroacetic acid, trichloroacetic acid, fluoroacetic acid, difluoroacetic acid, and trifluoroacetic acid. In some embodiments, a deblocking agent removes one or more trityl groups from, for example, an oligomer, a support-bound oligomer, a support-bound subunit, or other protected nitrogen or oxygen moiety.

The terms "halogen" and "halo" refer to an atom selected from the group consisting of fluorine, chlorine, bromine, and iodine.

The term "capping agent" refers to a composition (e.g., a solution) comprising an acid anhydride (e.g., benzoic anhydride, acetic anhydride, phenoxyacetic anhydride, and the like) useful for blocking a reactive site of, for example, a support-medium forming a chemical bond with an incoming subunit or other activated group.

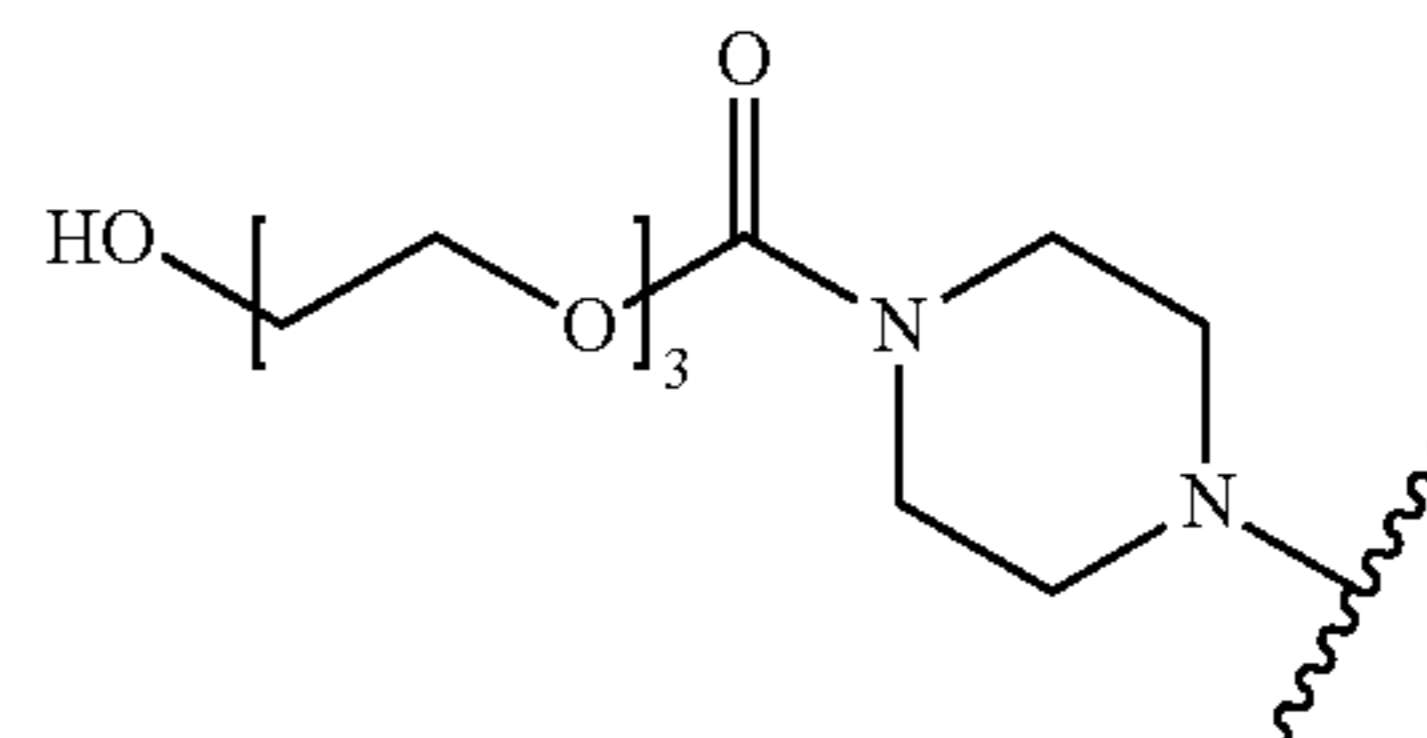
The term "cleavage agent" refers to a composition (e.g., a liquid solution or gaseous mixture) comprising a chemical base (e.g., ammonia or 1,8-diazabicycloundec-7-ene) or a combination of chemical bases useful for cleaving, for example, a support-bound oligomer from a support-medium.

The term "deprotecting agent" refers to a composition (e.g., a liquid solution or gaseous mixture) comprising a chemical base (e.g., ammonia, 1,8-diazabicycloundec-7-ene or potassium carbonate) or a combination of chemical bases useful for removing protecting groups. For example, a deprotecting agent, in some embodiments, can remove the base protection from, for example, a morpholino subunit, morpholino subunits of a morpholino oligomer, or support-bound versions thereof.

The term "solvent" refers to a component of a solution or mixture in which a solute is dissolved. Solvents may be inorganic or organic (e.g., acetic acid, acetone, acetonitrile, acetyl acetone, 2-aminoethanol, aniline, anisole, benzene, benzonitrile, benzyl alcohol, 1-butanol, 2-butanol, i-butanol, 2-butanone, t-butyl alcohol, carbon disulfide, carbon tetrachloride, chlorobenzene, chloroform, cyclohexane, cyclohexanol, cyclohexanone, di-n-butylphthalate, 1,1-dichloroethane, 1,2-dichloroethane, diethylamine, diethylene glycol, diglyme, dimethoxyethane (glyme), N,N-dimethylaniline, dimethylformamide, dimethylphthalate, dimethylsulfoxide, dioxane, ethanol, ether, ethyl acetate, ethyl acetoacetate, ethyl benzoate, ethylene glycol, glycerin, heptane, 1-heptanol, hexane, 1-hexanol, methanol, methyl acetate, methyl t-butyl ether, methylene chloride, 1-octanol, pentane, 1-pentanol, 2-pentanol, 3-pentanol, 2-pentanone, 3-pentanone, 1-propanol, 2-propanol, pyridine, tetrahydrofuran, toluene, water, p-xylene).

The phrases "morpholino oligomer" and "phosphorodiamidate morpholino oligomer" or "PMO" refers to an oligomer having morpholino subunits linked together by phosphorodiamidate linkages, joining the morpholino nitrogen of one subunit to the 5'-exocyclic carbon of an adjacent subunit. Each morpholino subunit comprises a nucleobase-pairing moiety effective to bind, by nucleobase-specific hydrogen bonding, to a nucleobase in a target.

The term "EG3 tail" refers to triethylene glycol moieties conjugated to the oligomer, e.g., at its 3'- or 5'-end. For example, in some embodiments, "EG3 tail" conjugated to the 3' end of an oligomer can be of the structure:



In certain embodiments, an oligomeric compound prepared by the methods of the disclosure can have a length from 10 to 50 nucleotides, 10 to 40 nucleotides, 10 to 30 nucleotides, 10 to 25 nucleotides, 10 to 24 nucleotides, 10 to 23 nucleotides, 10 to 22 nucleotides, 10 to 21 nucleotides, 10 to 20 nucleotides, 15 to 30 nucleotides, 15 to 25 nucleotides, 15 to 24 nucleotides, 15 to 23 nucleotides, 15 to 22 nucleotides, 15 to 21 nucleotides, 15 to 20 nucleotides, 20 to 30 nucleotides, 20 to 25 nucleotides, 20 to 24 nucleotides, 20 to 23 nucleotides, or 20 to 22 nucleotides in length, including all integers in between these ranges. Preferably, an oligomeric compound prepared by the methods of the disclosure is of 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 20 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 21 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 22 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 23 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 24 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 25 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 26 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the dis-

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closure is 27 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 28 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 29 nucleotides in length. In some embodiments, an oligomeric compound prepared by the methods of the disclosure is 30 nucleotides in length.

Accordingly, in some embodiments of oligomeric compounds prepared by the methods of the disclosure including, for example, embodiments of oligomeric compounds of Formula (A), (B), (D), (G), (F), and (S), n is an integer ranging from 10 to 50, 10 to 40, 10 to 30, 10 to 25, 10 to 24, 10 to 23, 10 to 22, 10 to 21, 10 to 20, 15 to 30, 15 to 25, 15 to 24, 15 to 23, 15 to 22, 15 to 21, 15 to 20, 20 to 30, 20 to 25, 20 to 24, 20 to 23, or 20 to 22. In certain embodiments, n is 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40. In some embodiments, n is 10. In some embodiments, n is 11. In some embodiments, n is 12. In some embodiments, n is 15. In some embodiments, n is 18. In some embodiments, n is 20. In some embodiments, n is 21. In some embodiments, n is 22. In some embodiments, n is 23. In some embodiments, n is 24. In some embodiments, n is 25. In some embodiments, n is 26. In some embodiments, n is 27. In some embodiments, n is 28. In some embodiments, n is 29. In some embodiments, n is 30.

The terms "about" or "approximately" are generally understood by persons knowledgeable in the relevant subject area, but in certain circumstances can mean within 10%, or within $\pm 5\%$, of a given value or range.

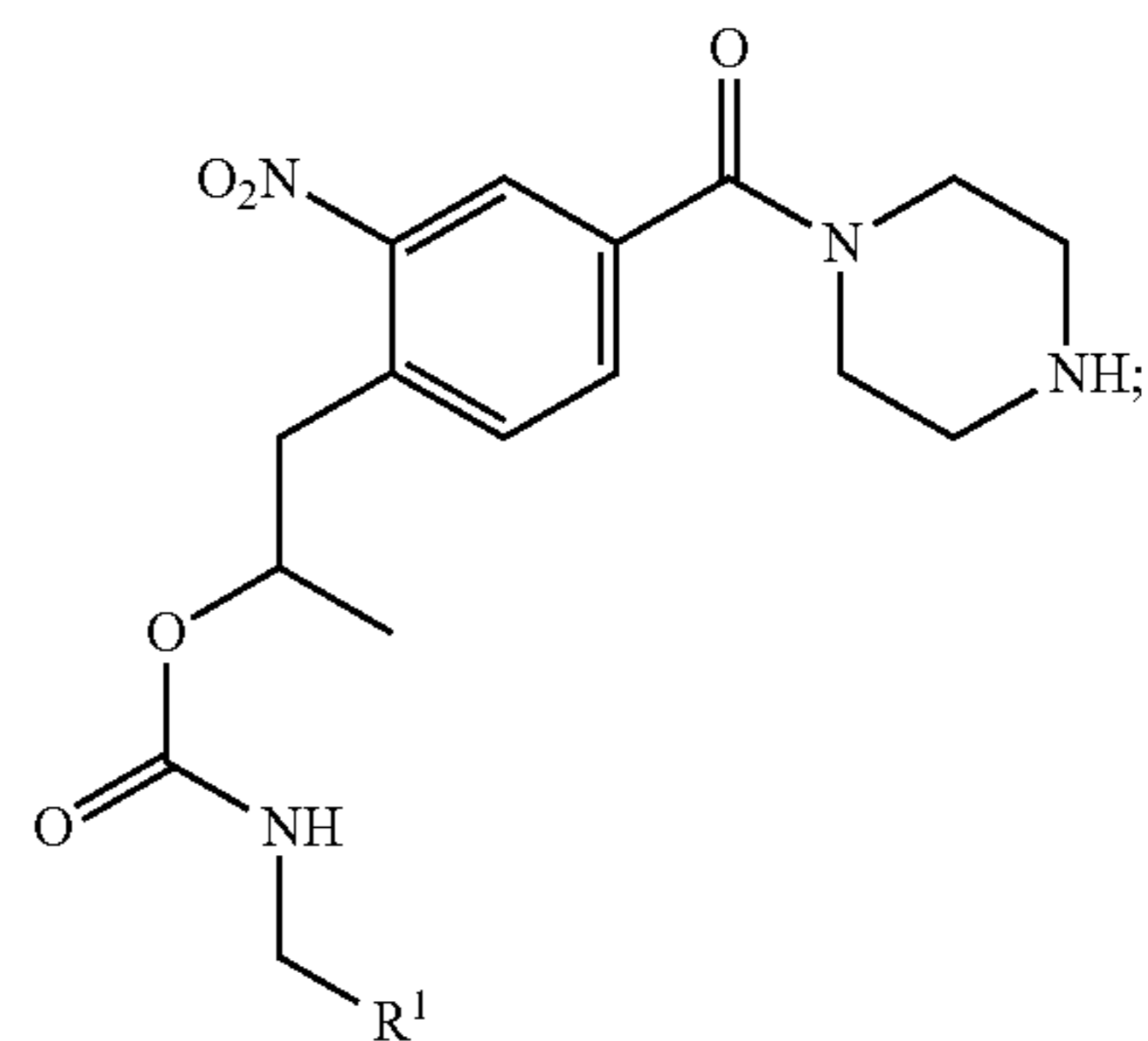
Processes for Preparing Morpholino Oligomers

Synthesis is generally prepared, as described herein, on a support-medium. In general a first synthon (e.g., a monomer, such as a morpholino subunit) is first attached to a support-medium, and the oligomer is then synthesized by sequentially coupling subunits to the support-bound synthon. This iterative elongation eventually results in a final oligomeric compound. Suitable support-media can be soluble or insoluble, or may possess variable solubility in different solvents to allow the growing support-bound polymer to be either in or out of solution as desired. Traditional support-media are for the most part insoluble and are routinely placed in reaction vessels while reagents and solvents react with and/or wash the growing chain until the oligomer has reached the target length, after which the oligomer is cleaved from the support, and, if necessary, further worked up to produce the final polymeric compound. More recent approaches have introduced soluble supports including soluble polymer supports to allow precipitating and dissolving the iteratively synthesized product at desired points in the synthesis (Gravert et al., *Chem. Rev.* 1997, 97,489-510).

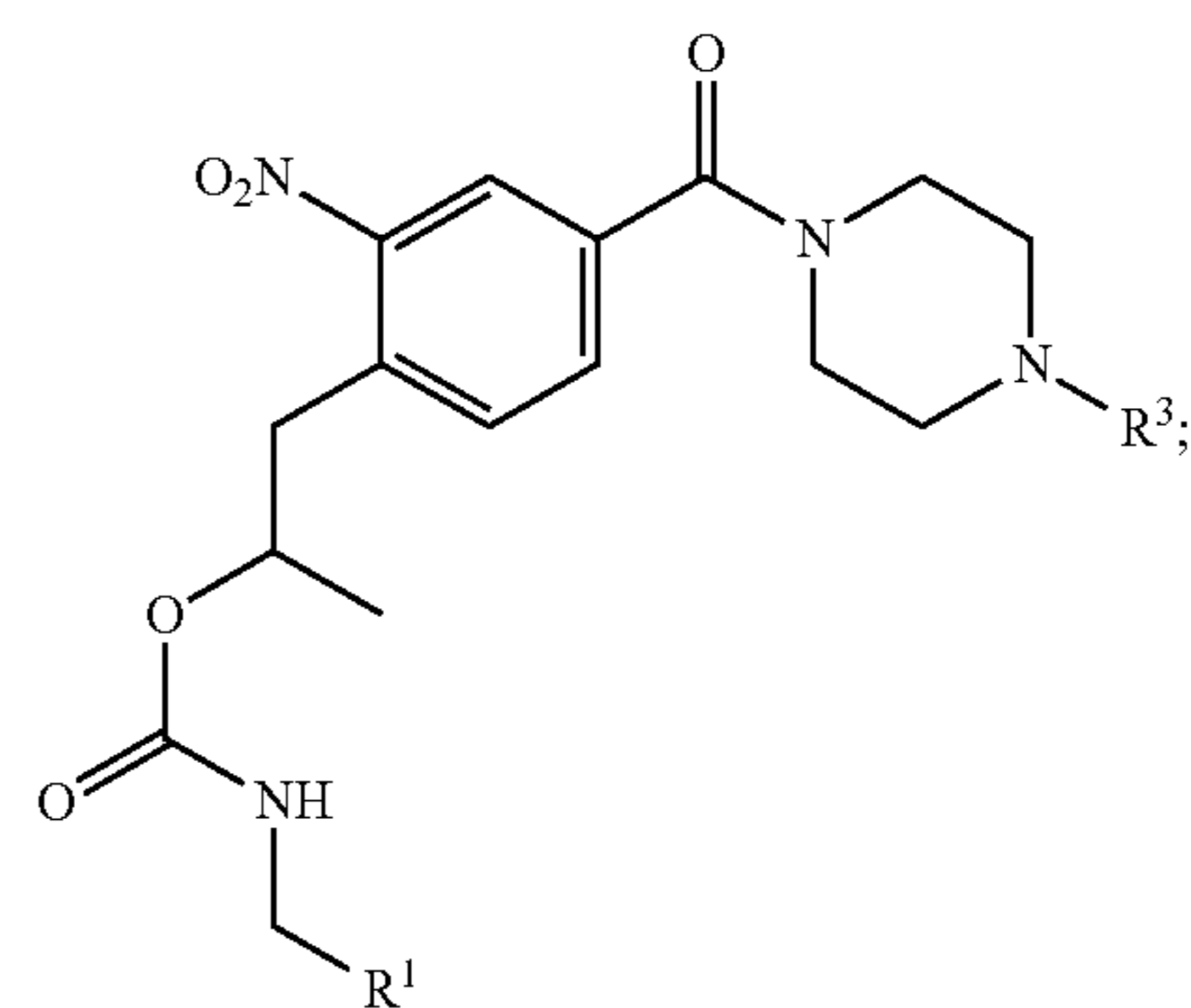
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Provided herein are processes for preparing morpholino oligomers).

Thus, in one aspect, provided herein is a process for preparing a compound of Formula (II):

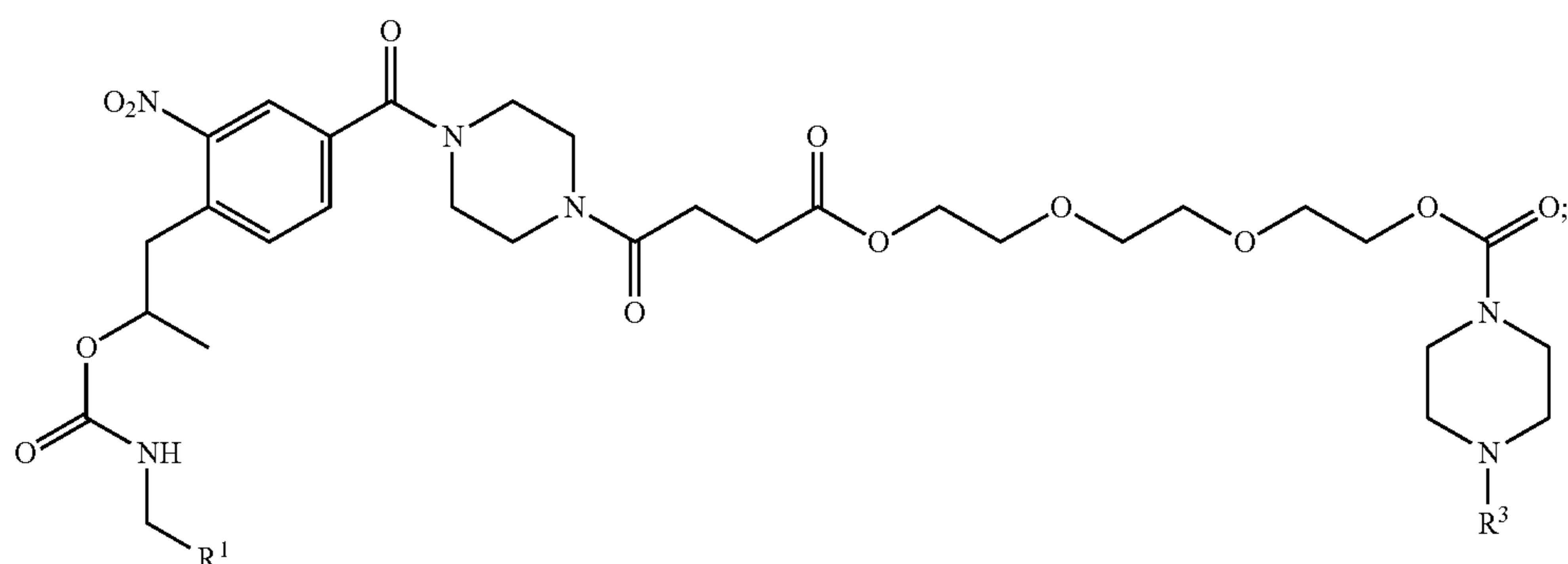


wherein R^1 is a support-medium;
wherein the process comprises contacting a compound of Formula (A1):



wherein R^1 is a support-medium and R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
with a deblocking agent to form the compound of Formula (II).

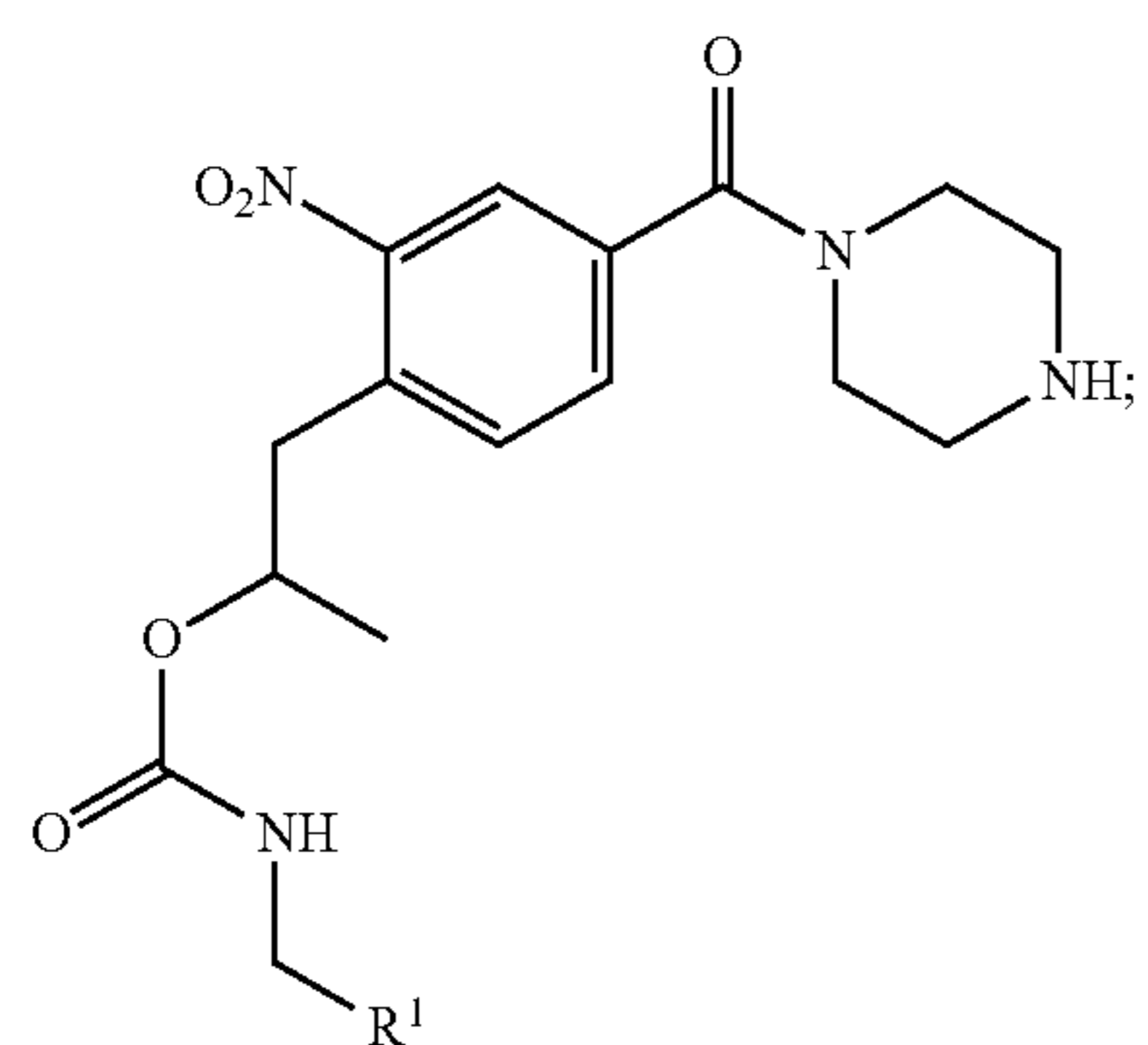
In another aspect, provided herein is a process for preparing a compound of Formula (A3):



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wherein R^1 is a support-medium, and R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
 wherein the process comprises contacting a compound of Formula (II):



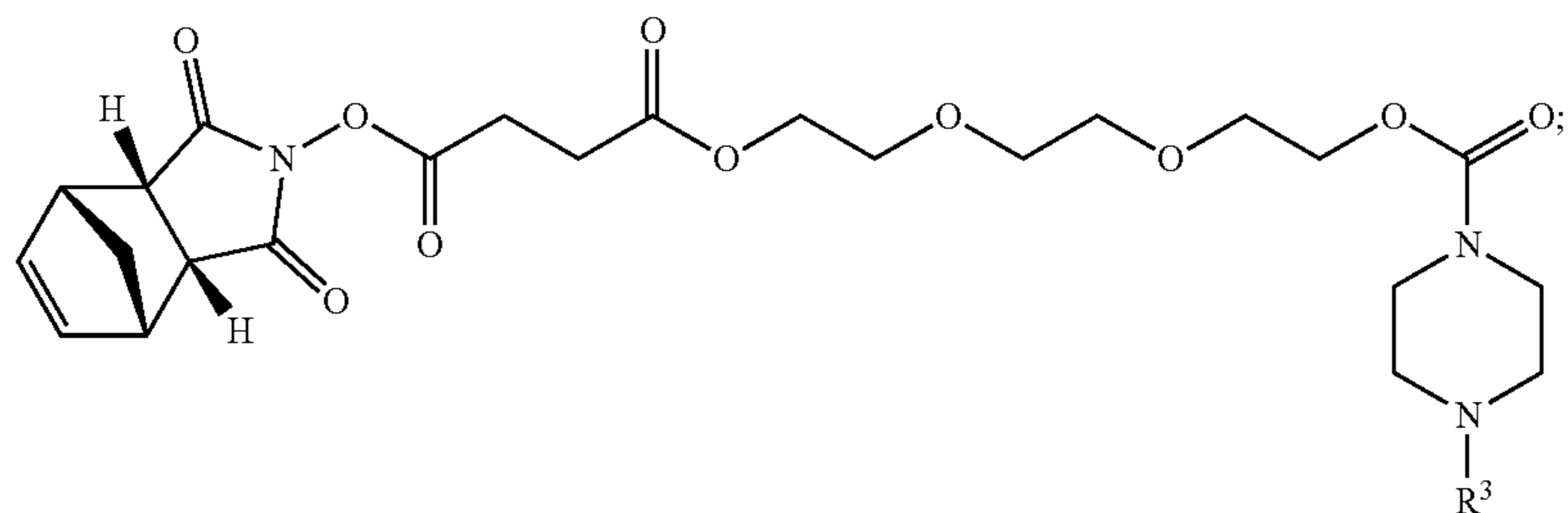
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wherein R^1 is a support-medium;
 with a compound of Formula (A2):

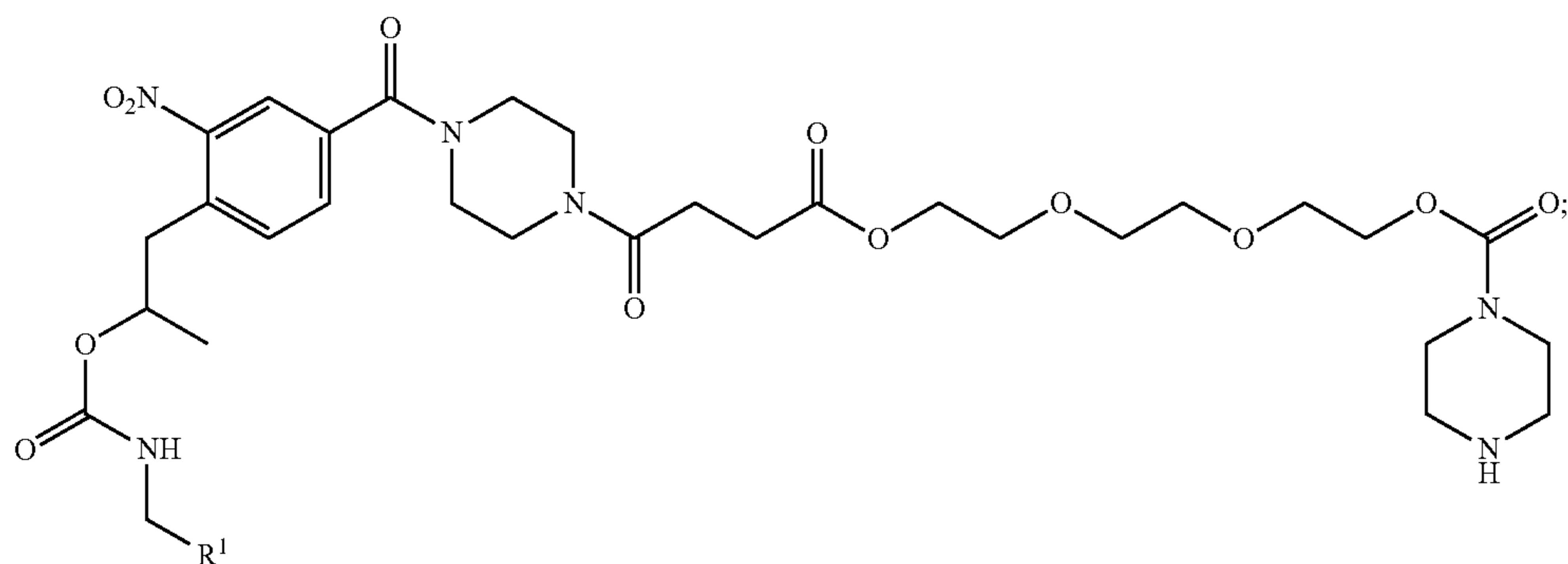


wherein R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

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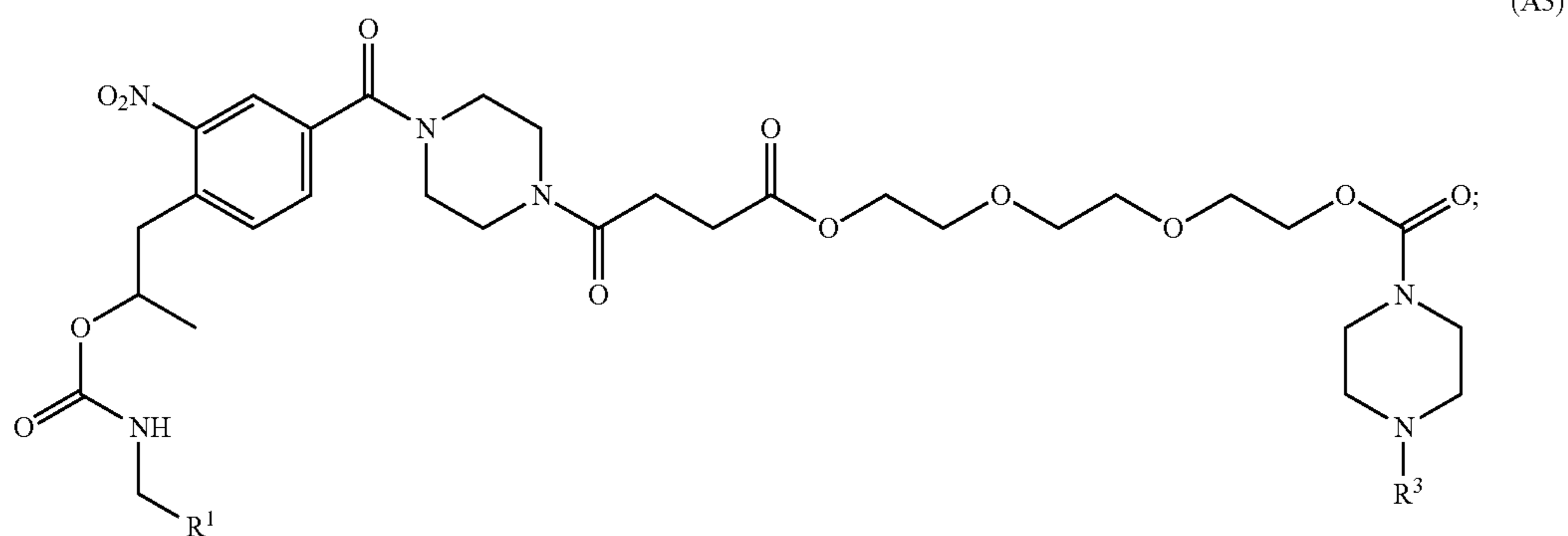
to form the compound of Formula (A3).

In still another aspect, provided herein is a process for preparing a compound of Formula (IV):



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wherein R^1 is a support-medium;
wherein the process comprises contacting a compound of
Formula (A3):

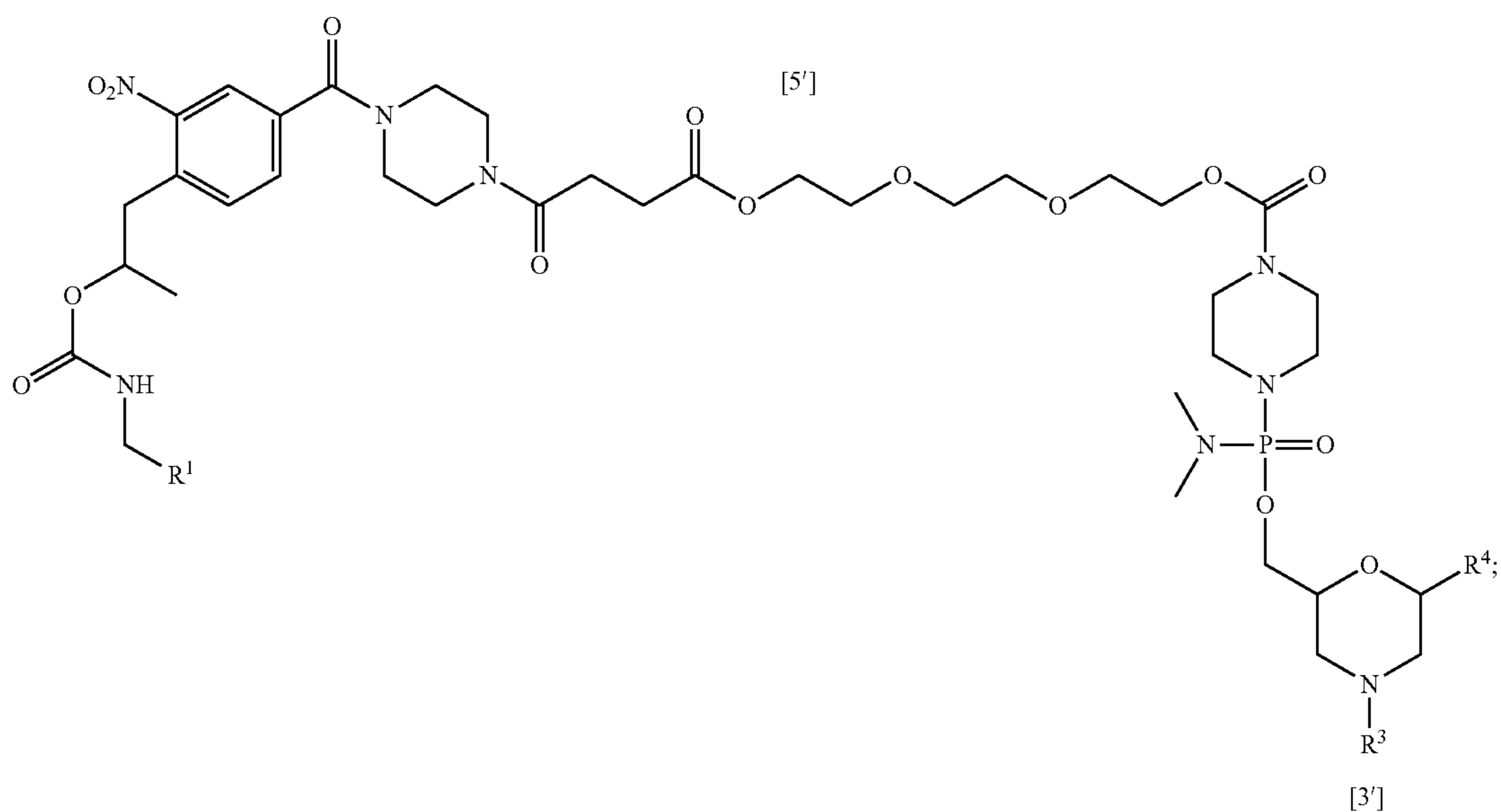


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wherein R^1 is a support-medium, and R^3 is selected from
the group consisting of trityl, monomethoxytrityl,
dimethoxytrityl and trimethoxytrityl;
with a deblocking agent to form a compound of Formula
(IV).

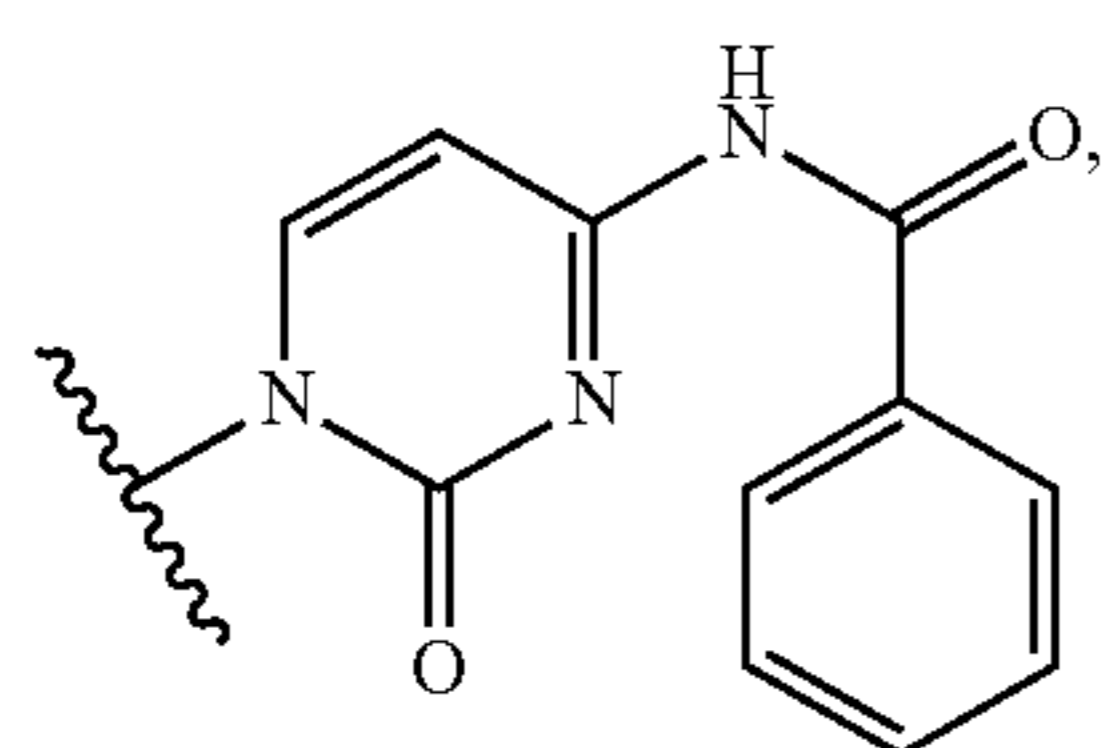
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In yet another aspect, provided herein is a process for
preparing a compound of Formula (A5):



wherein R^1 is a support-medium, R^3 is selected from the
group consisting of trityl, monomethoxytrityl, dimethoxytri-
tyl and trimethoxytrityl, and
 R^4 is selected from the group consisting of:

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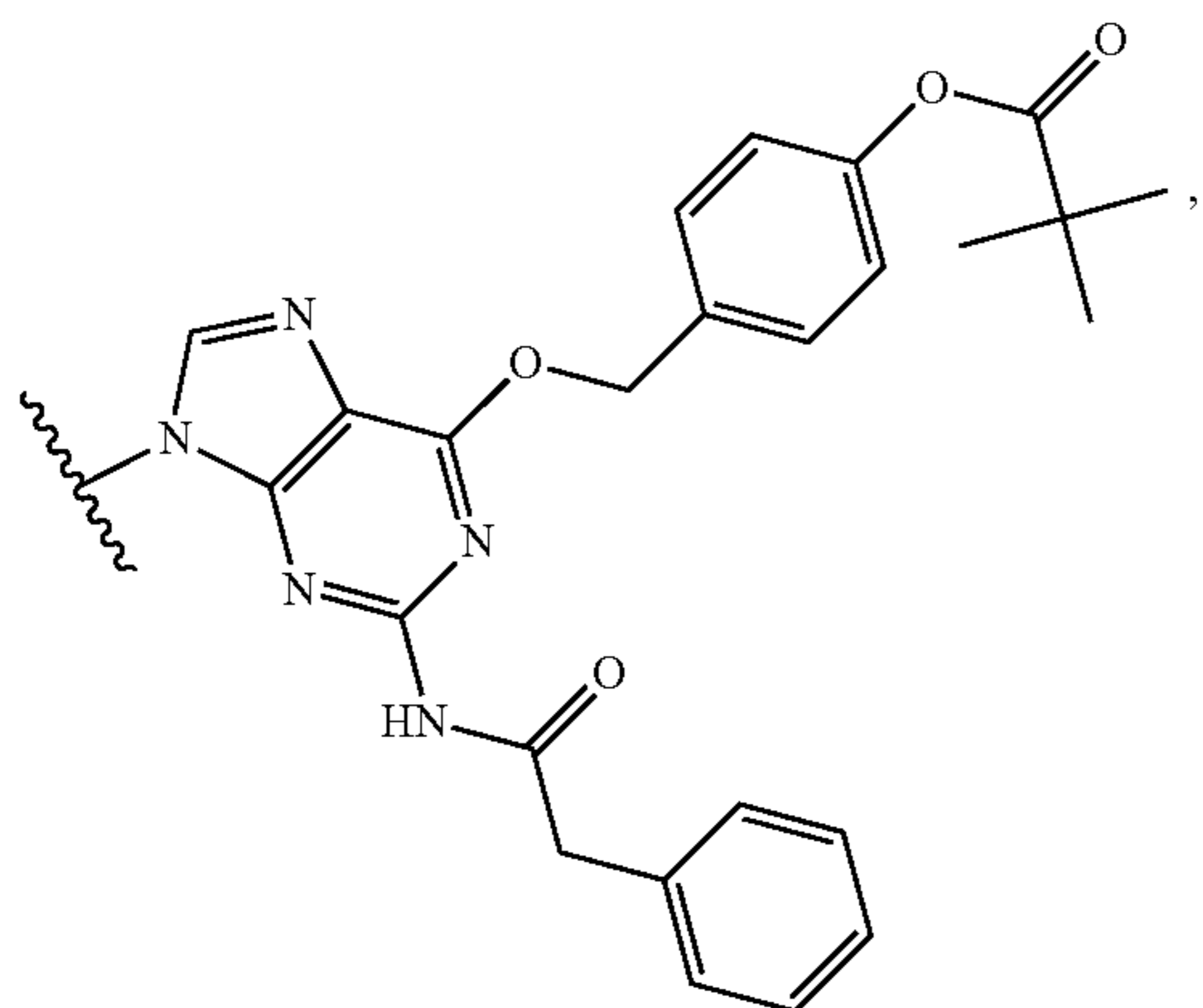


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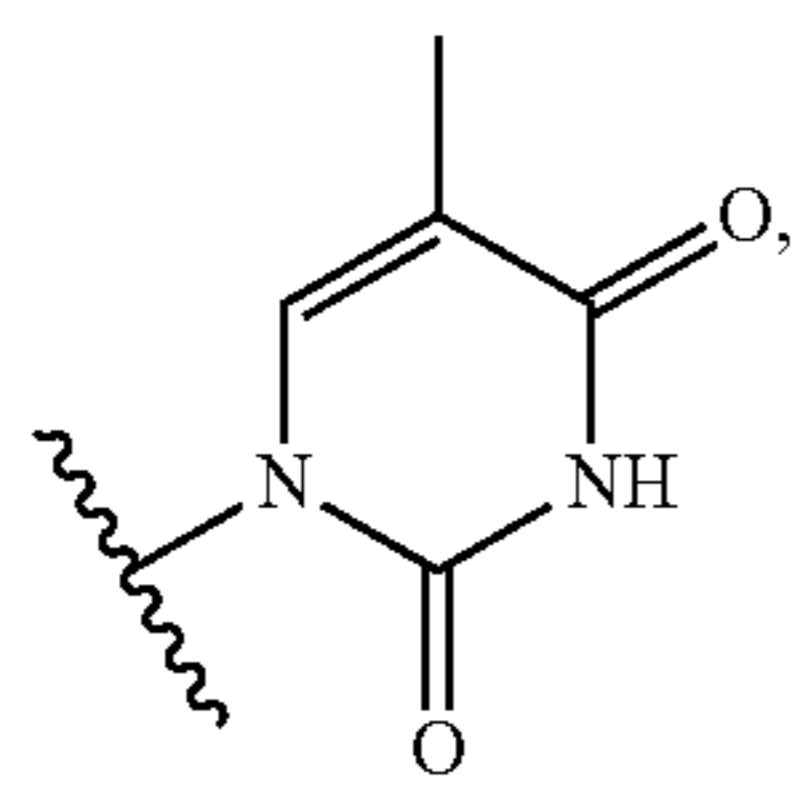


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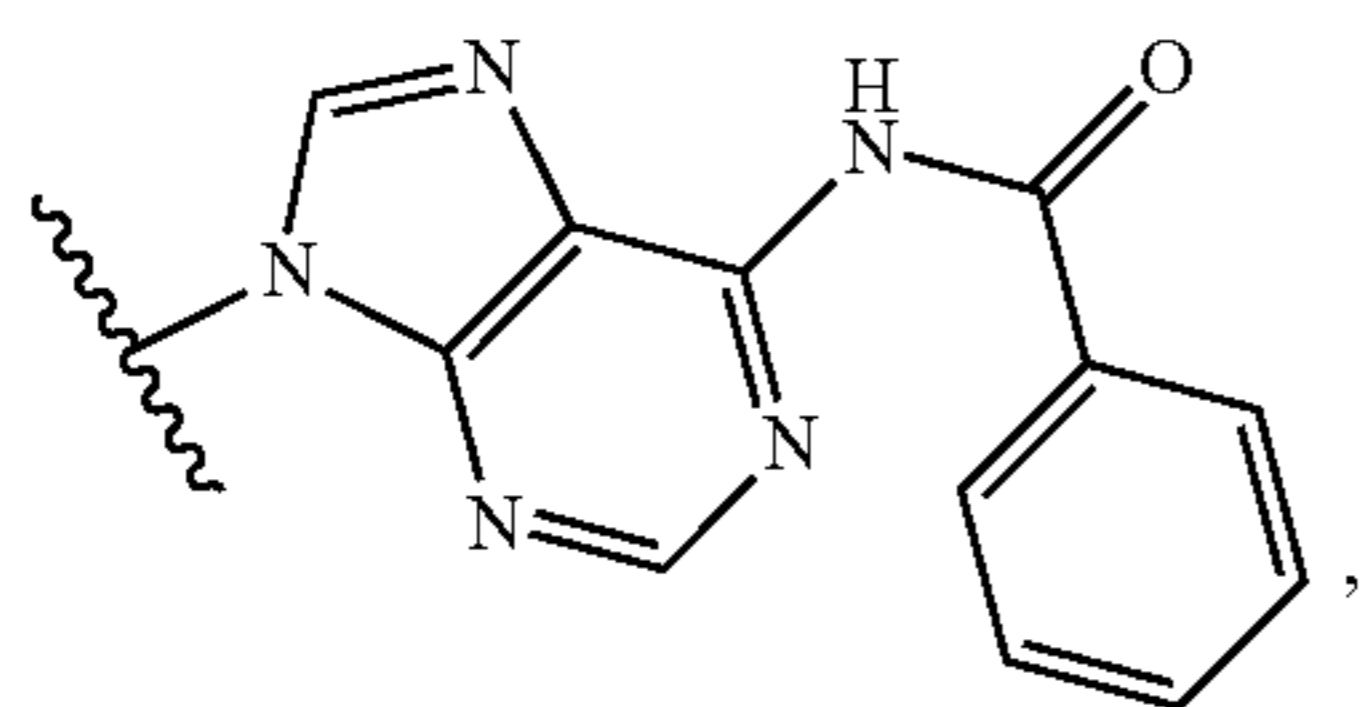
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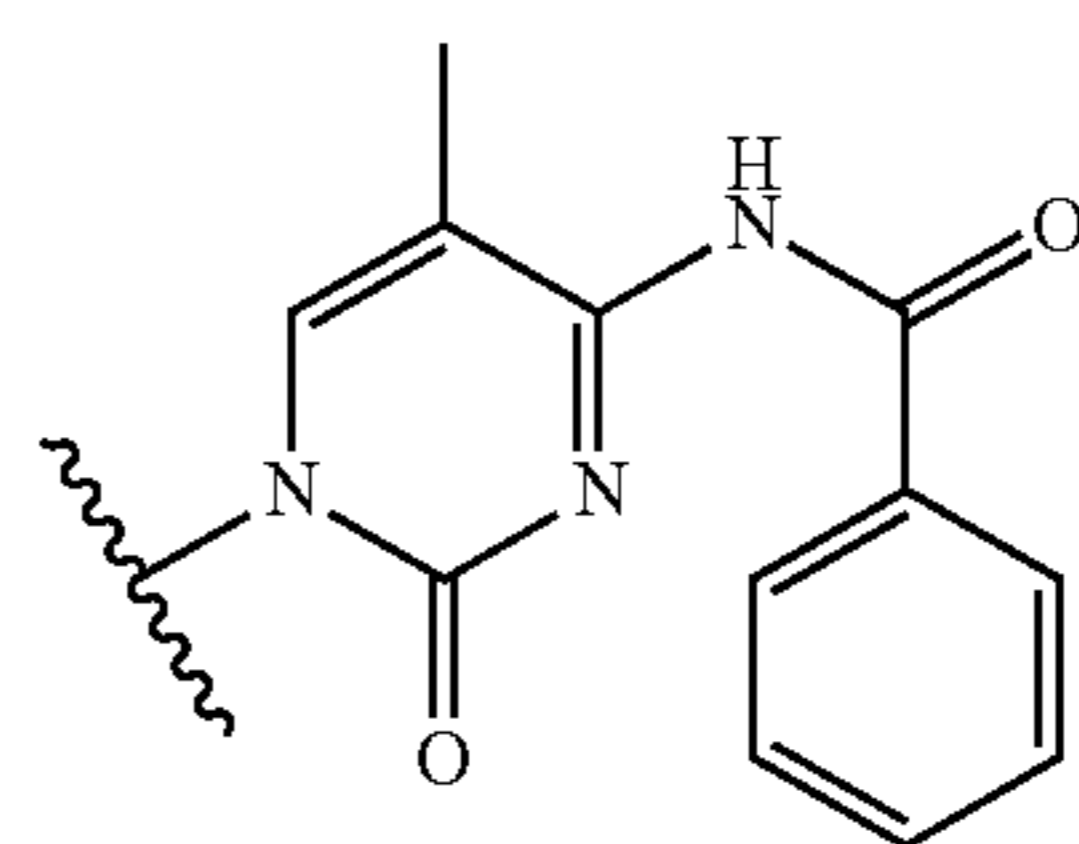


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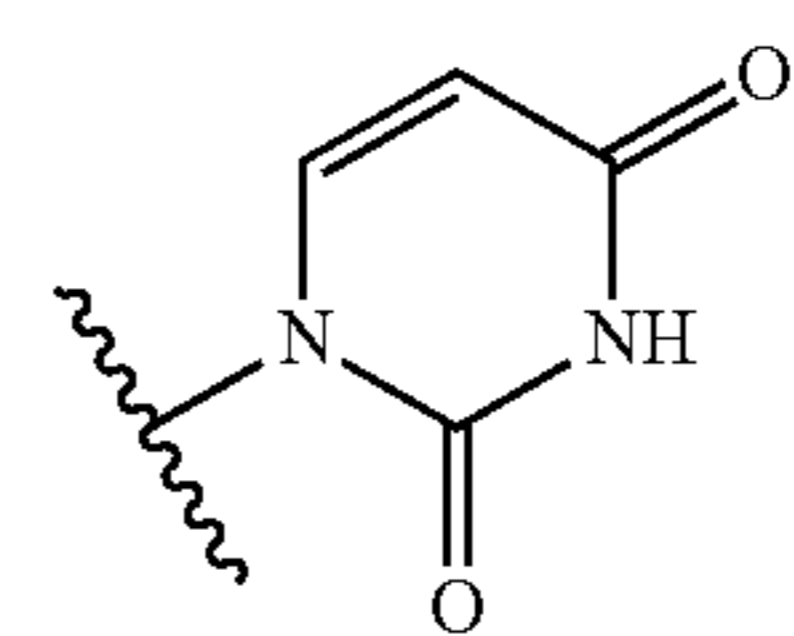
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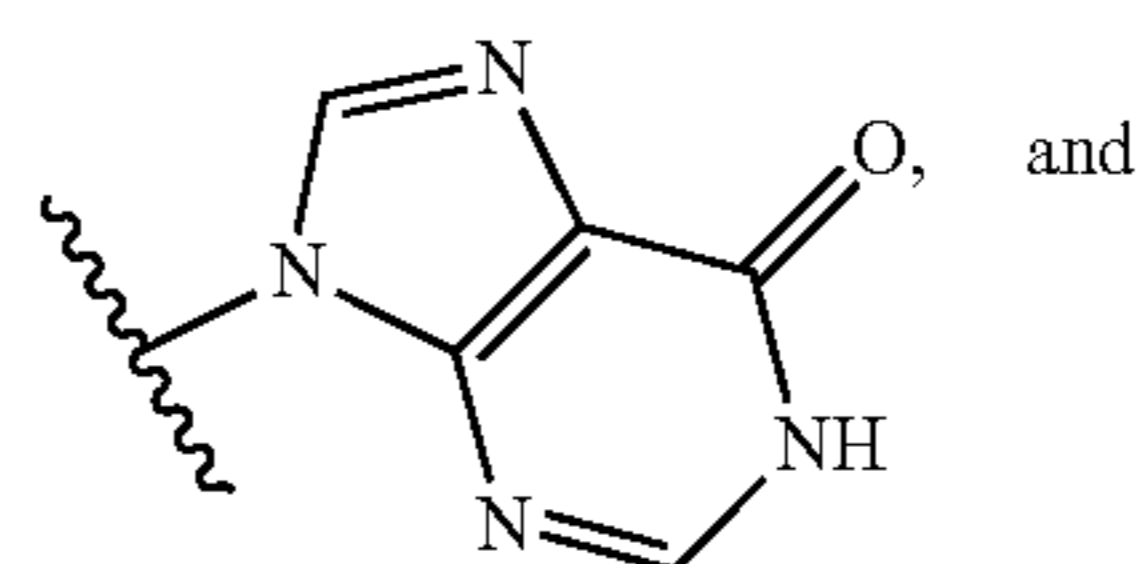


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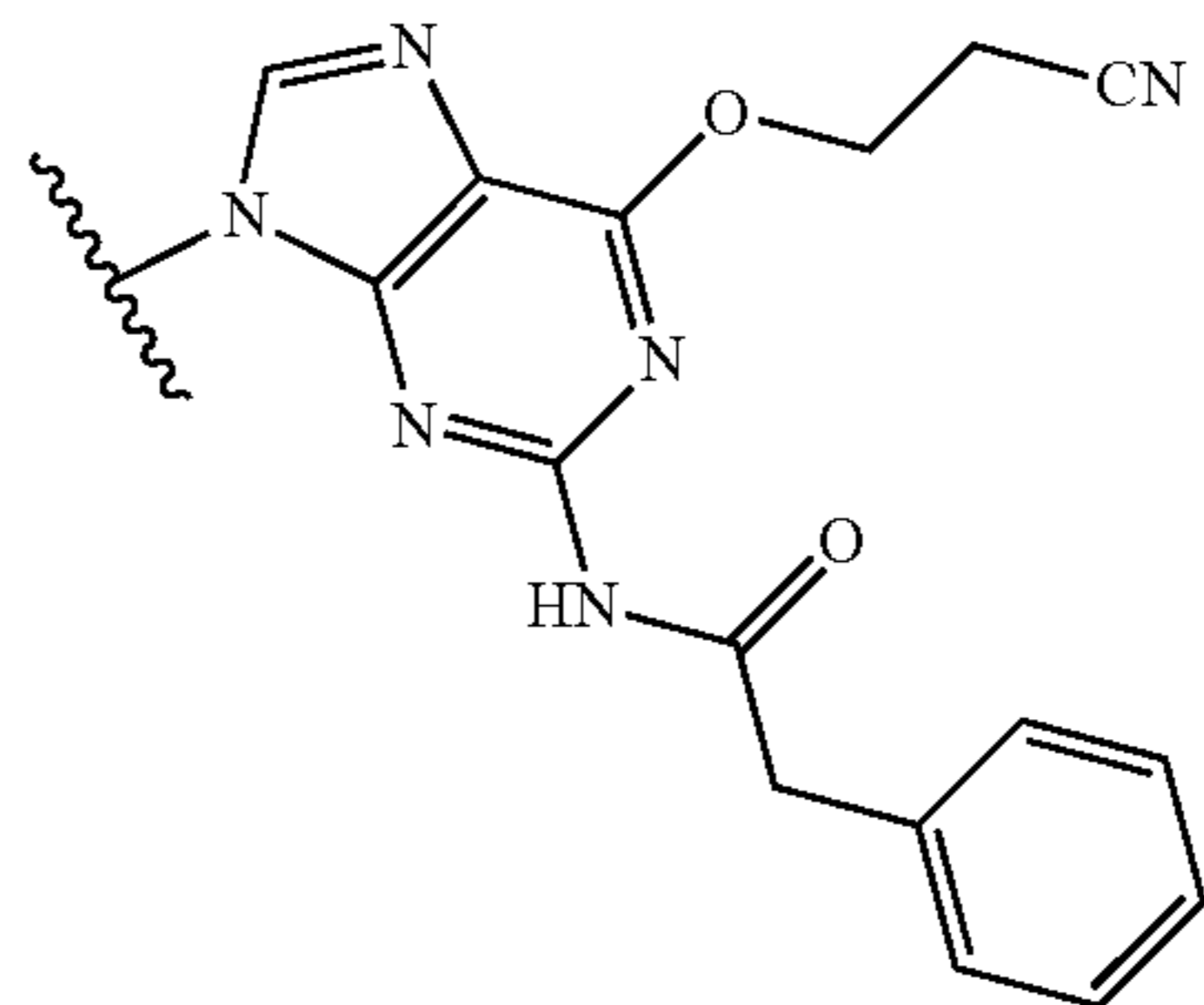
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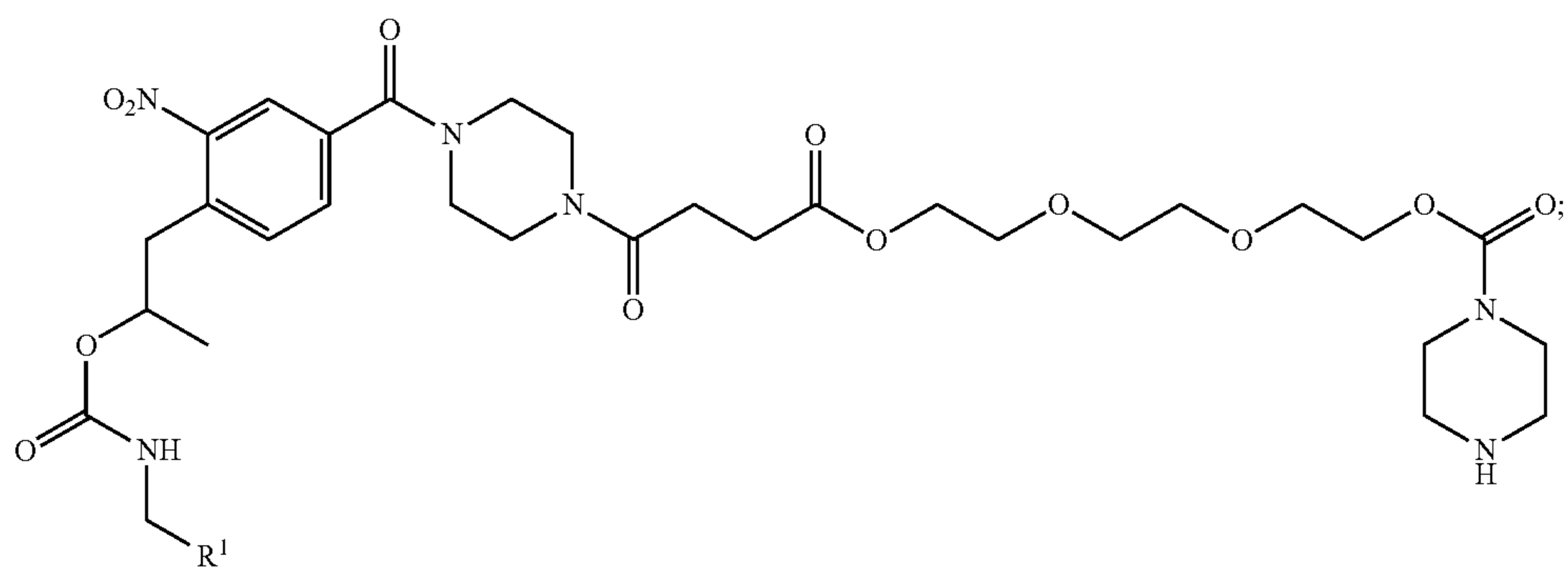
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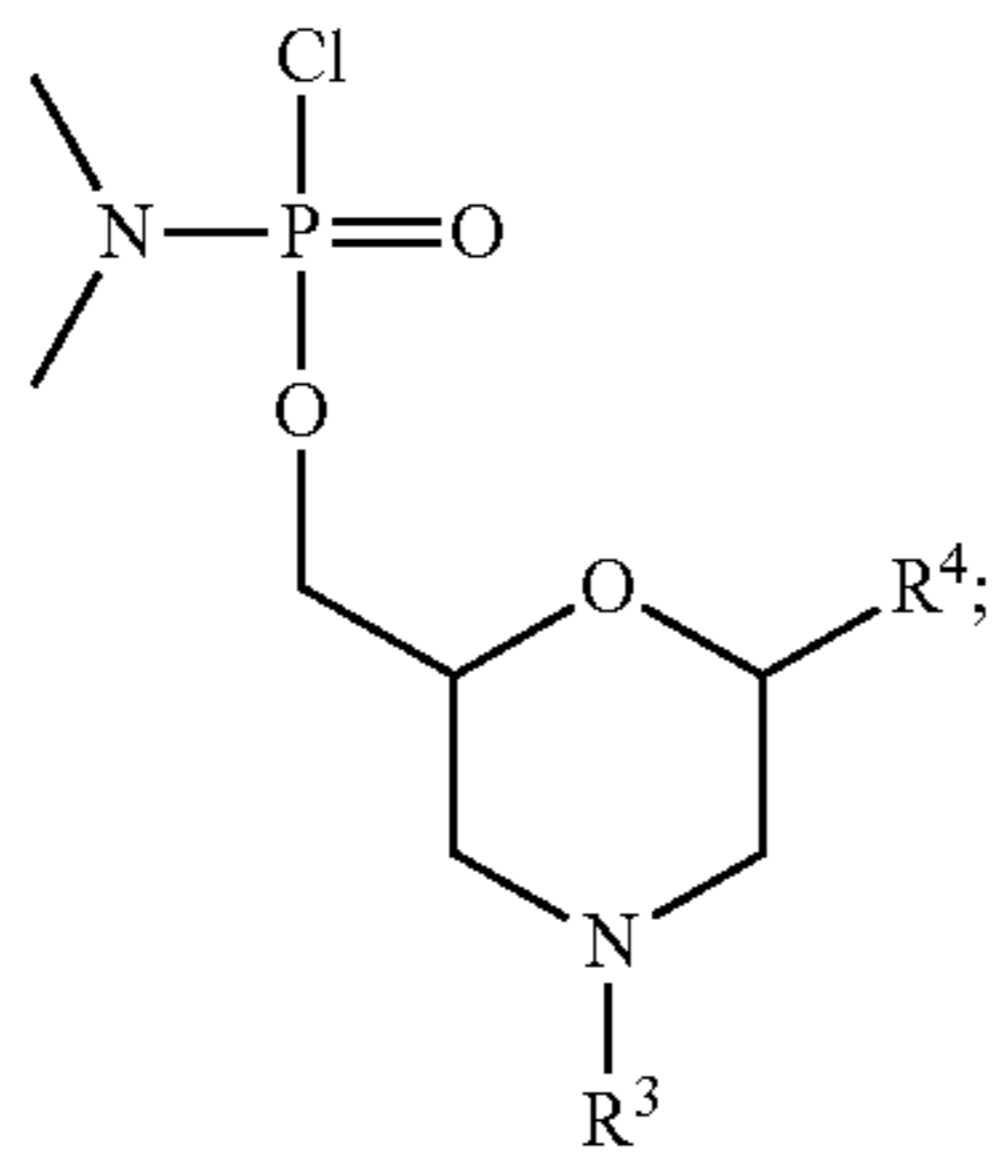


wherein the process comprises contacting a compound of Formula (IV):

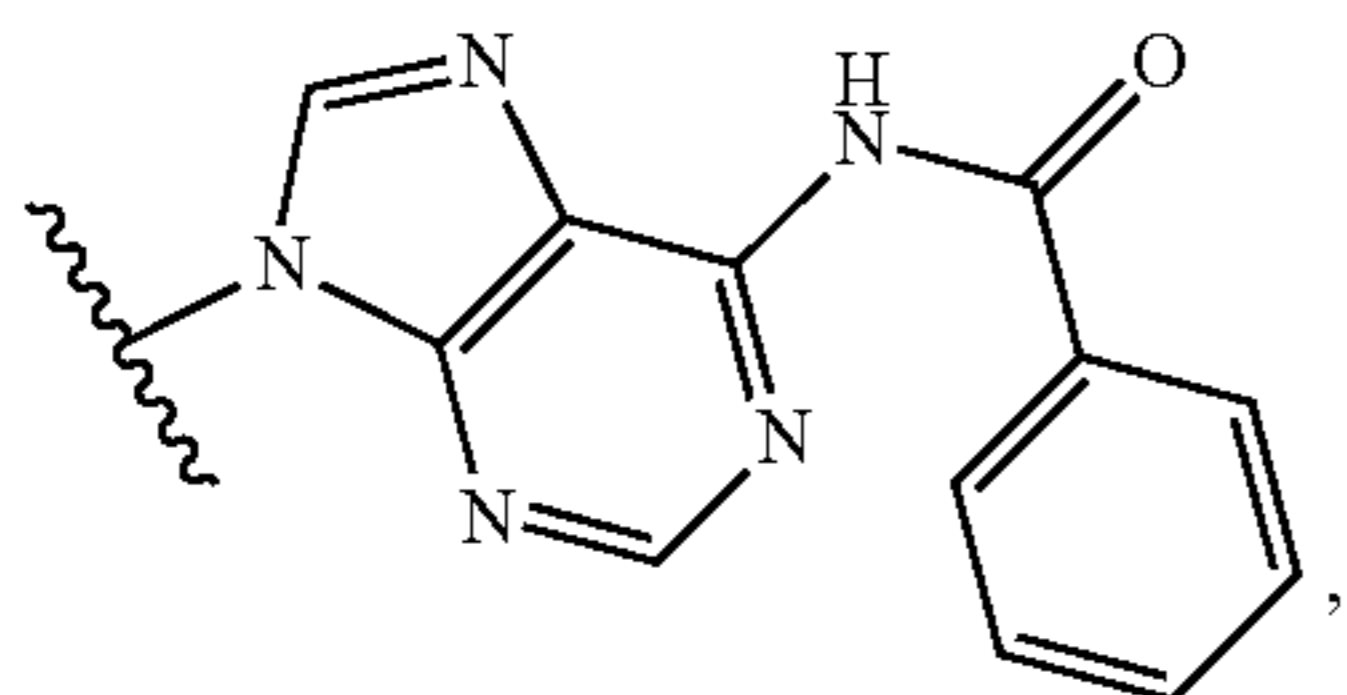
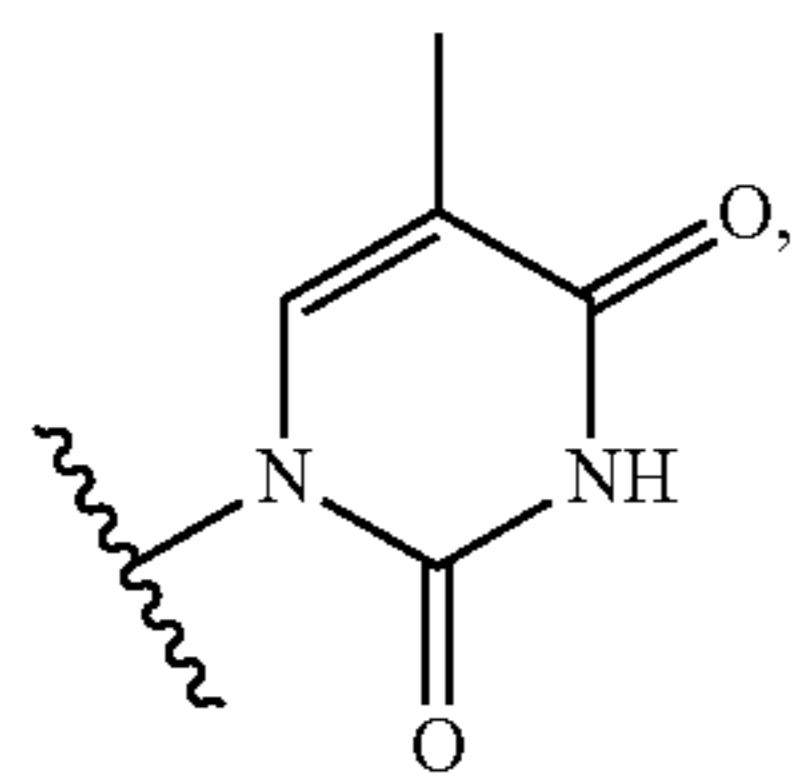
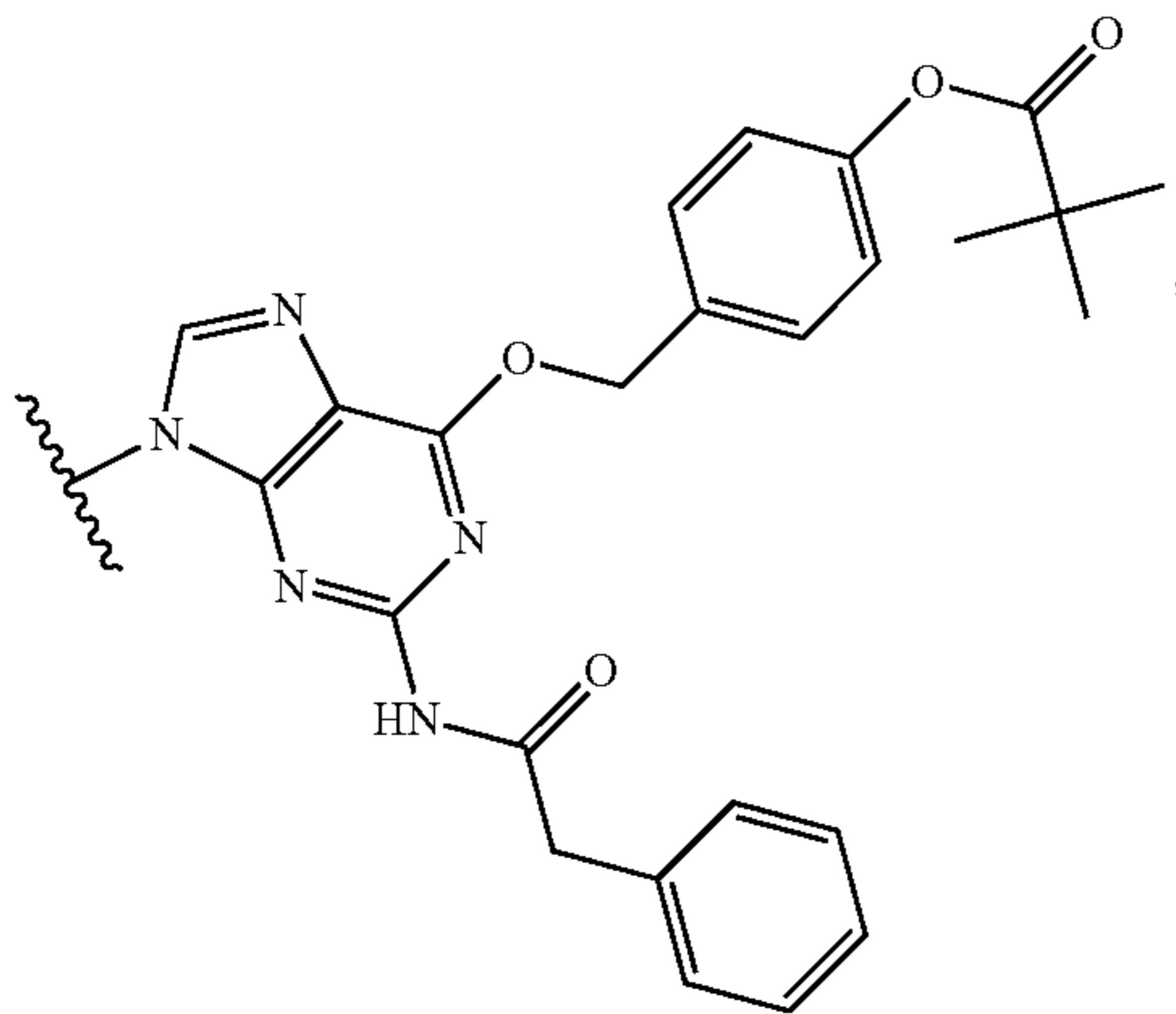
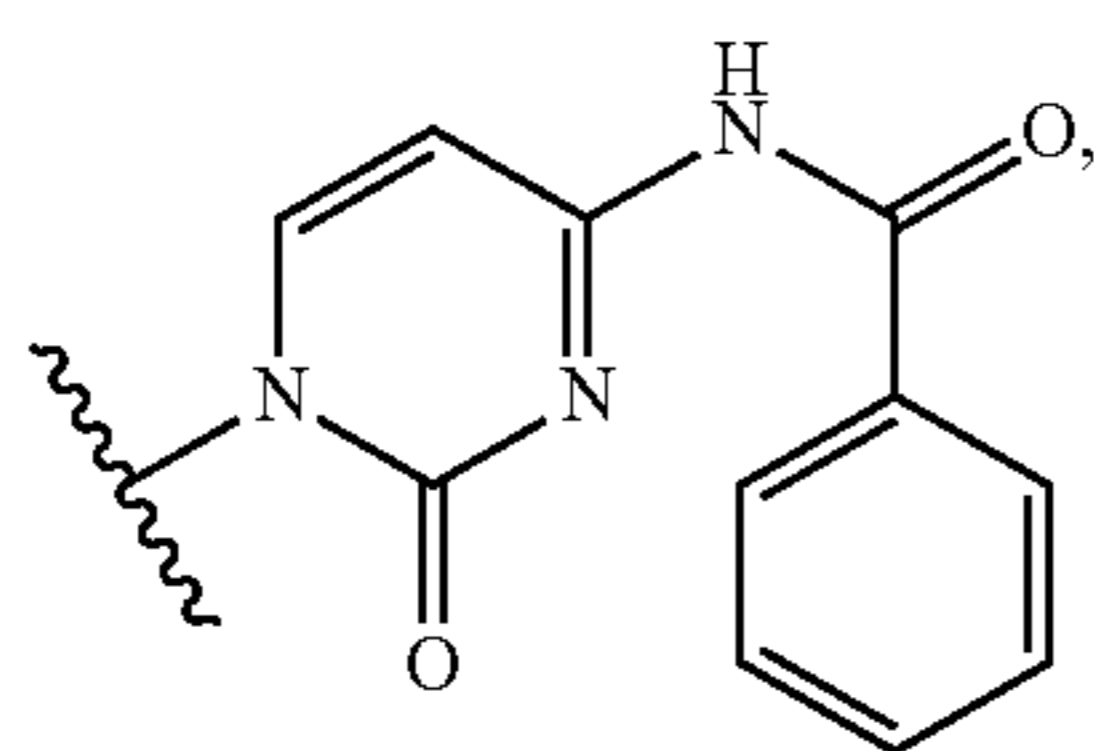


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wherein R¹ is a support-medium;
with a compound of Formula (A4):



wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

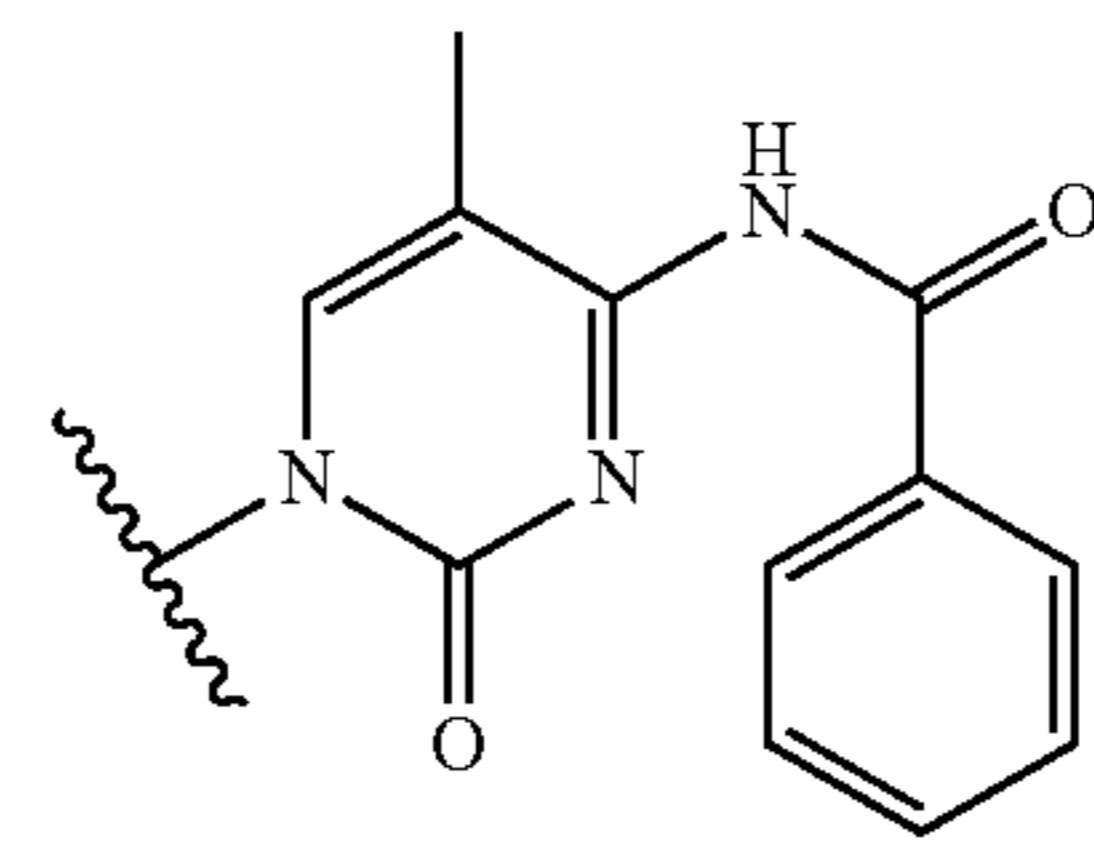


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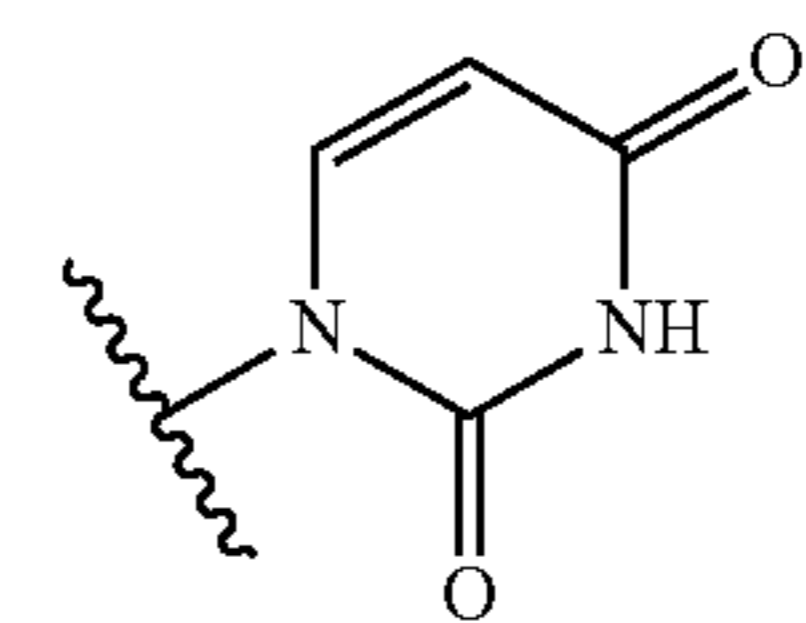
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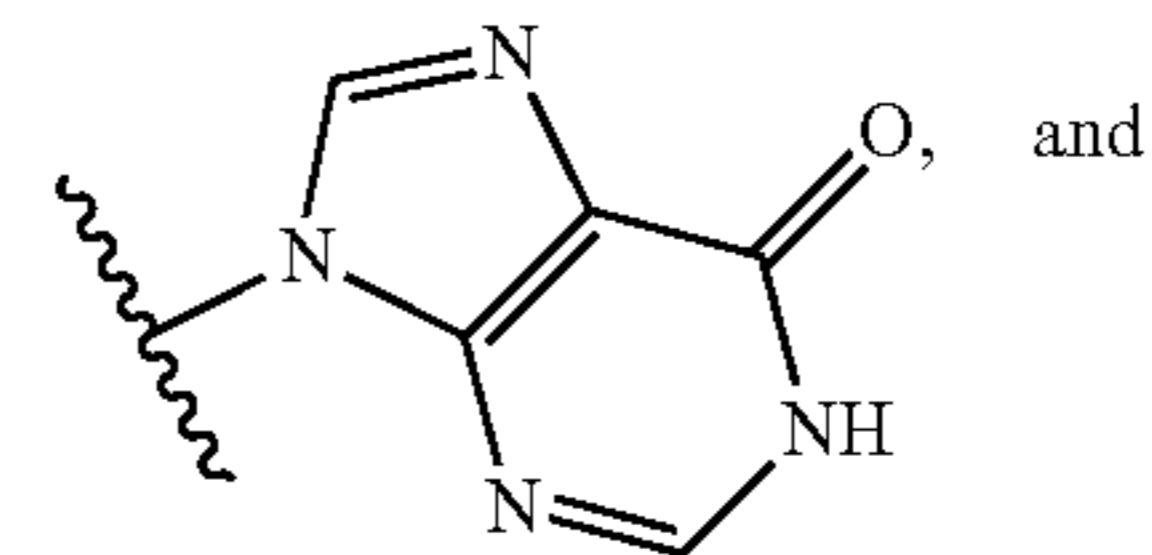
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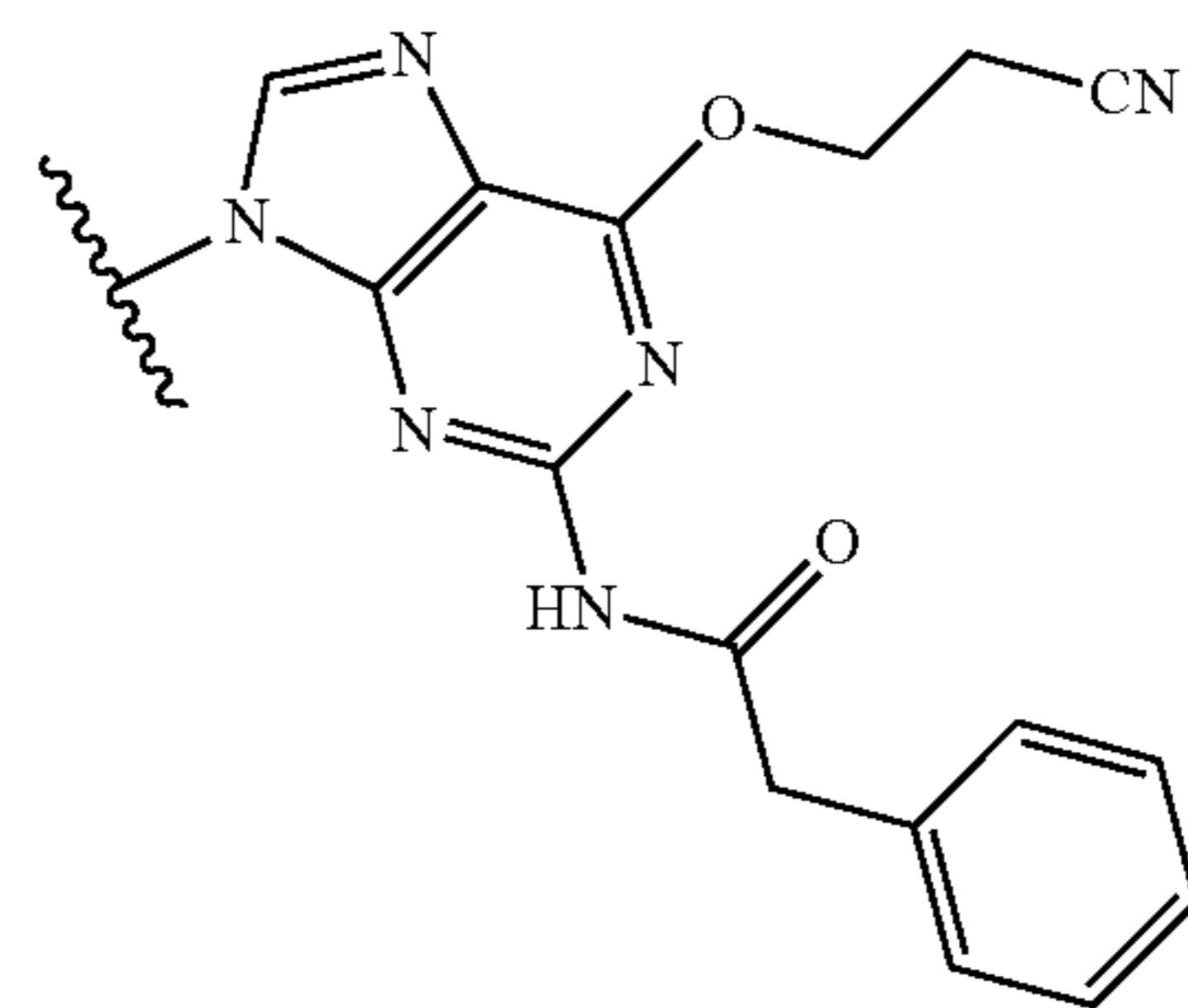
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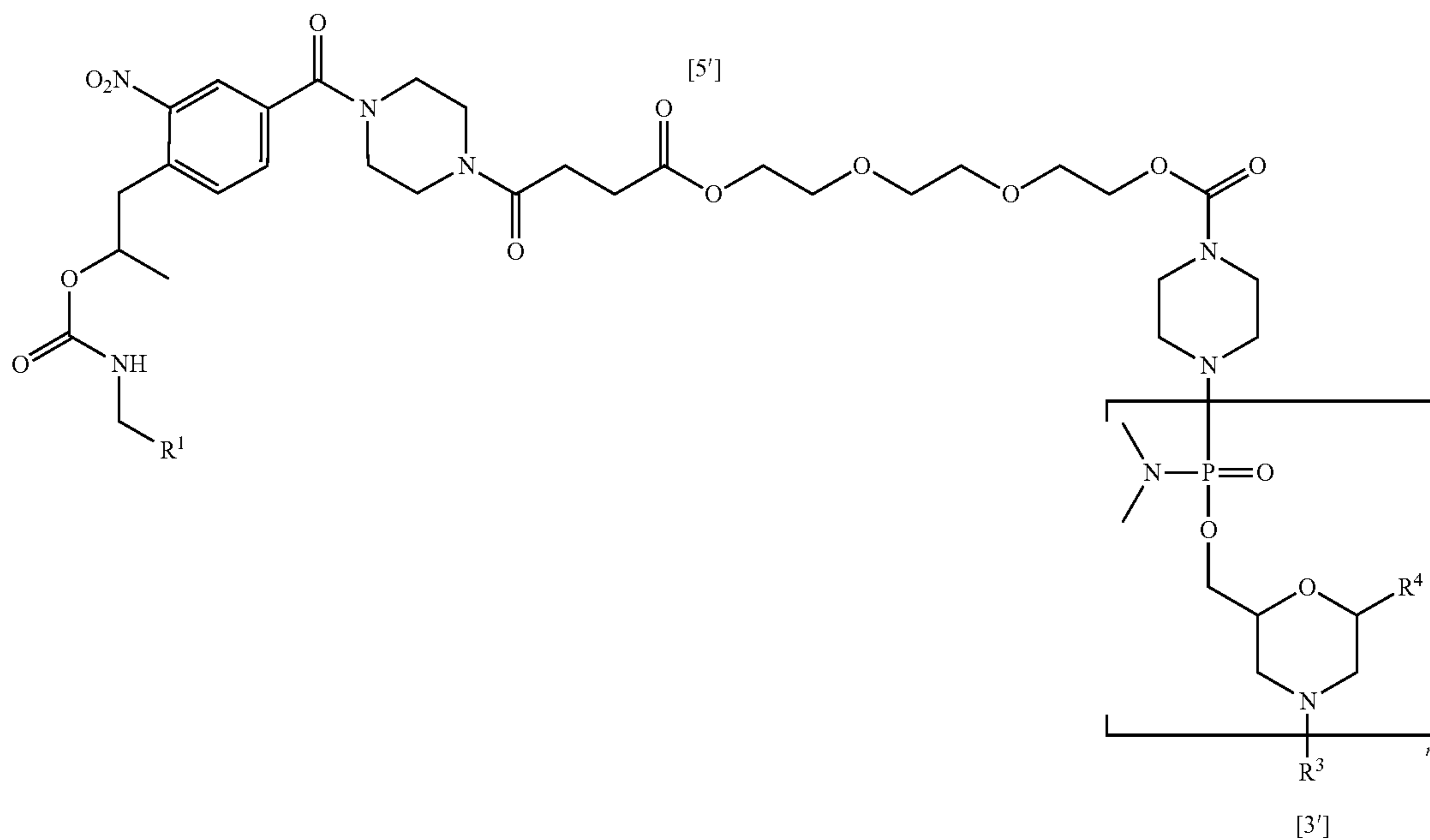
to form a compound of Formula (A5).

In another aspect, provided herein is a process for preparing a compound of Formula (A9):

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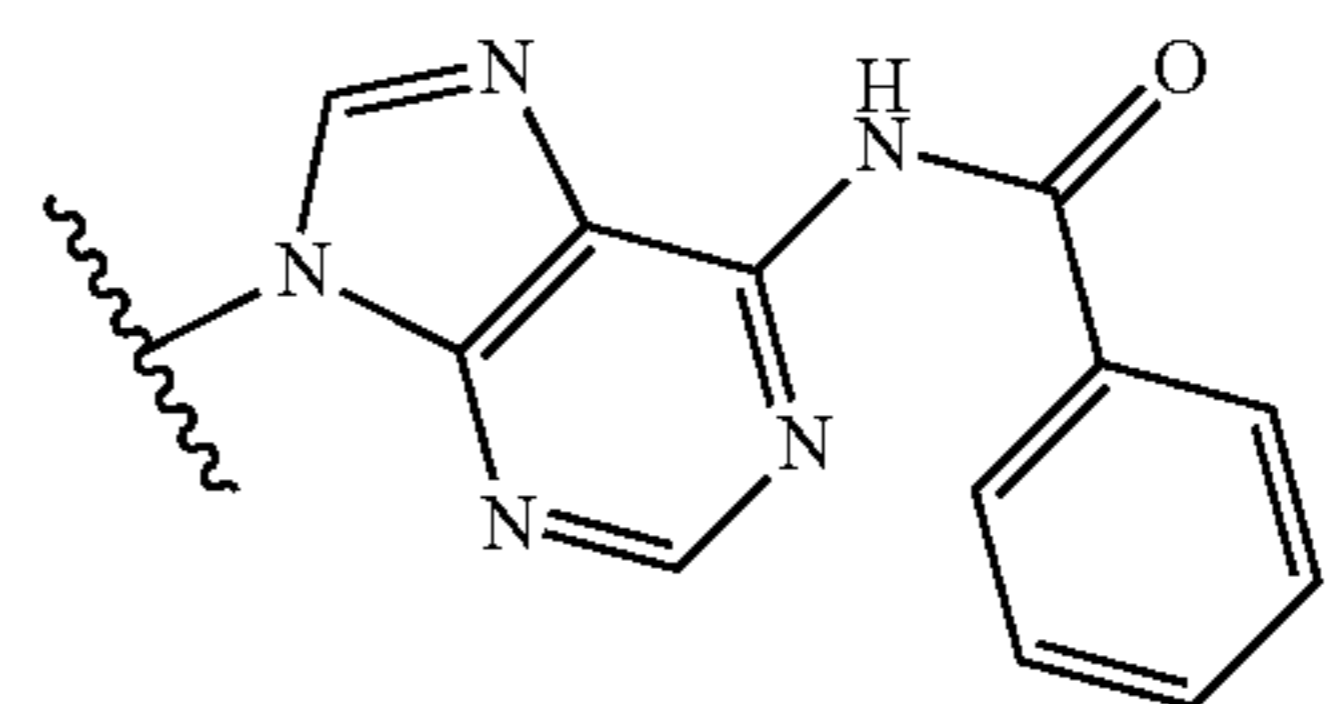
(A9)



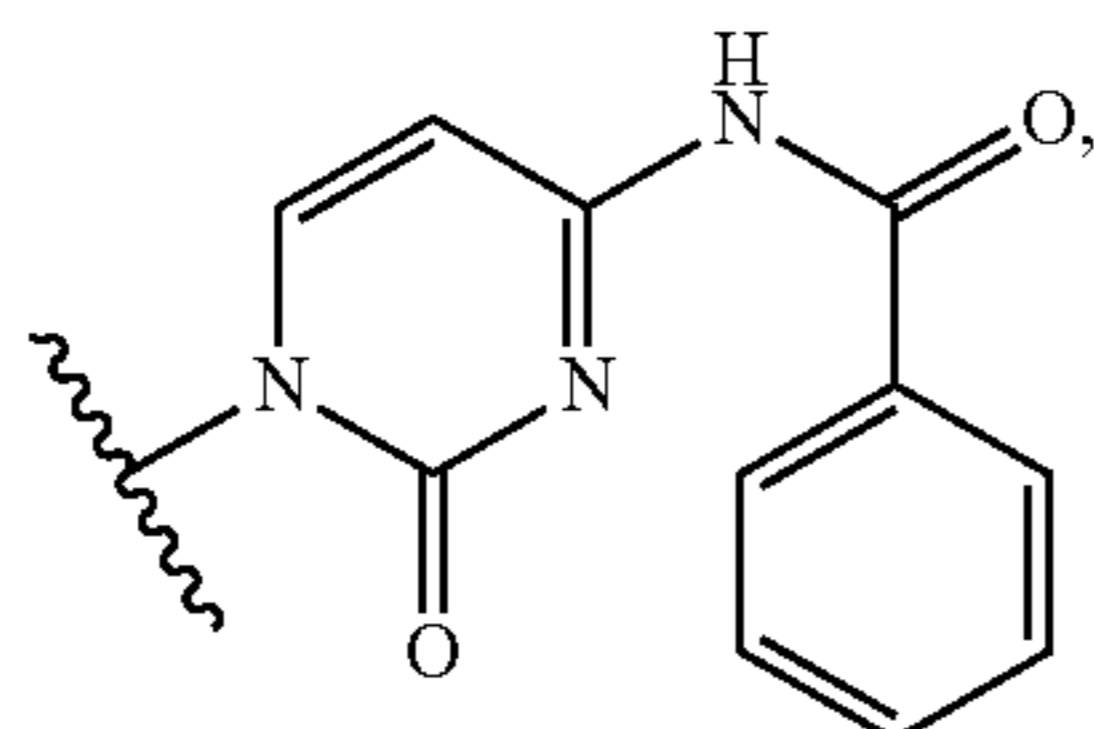
wherein n is an integer from 10 to 40, R¹ is a support-medium, R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each occurrence, selected from the group consisting of:

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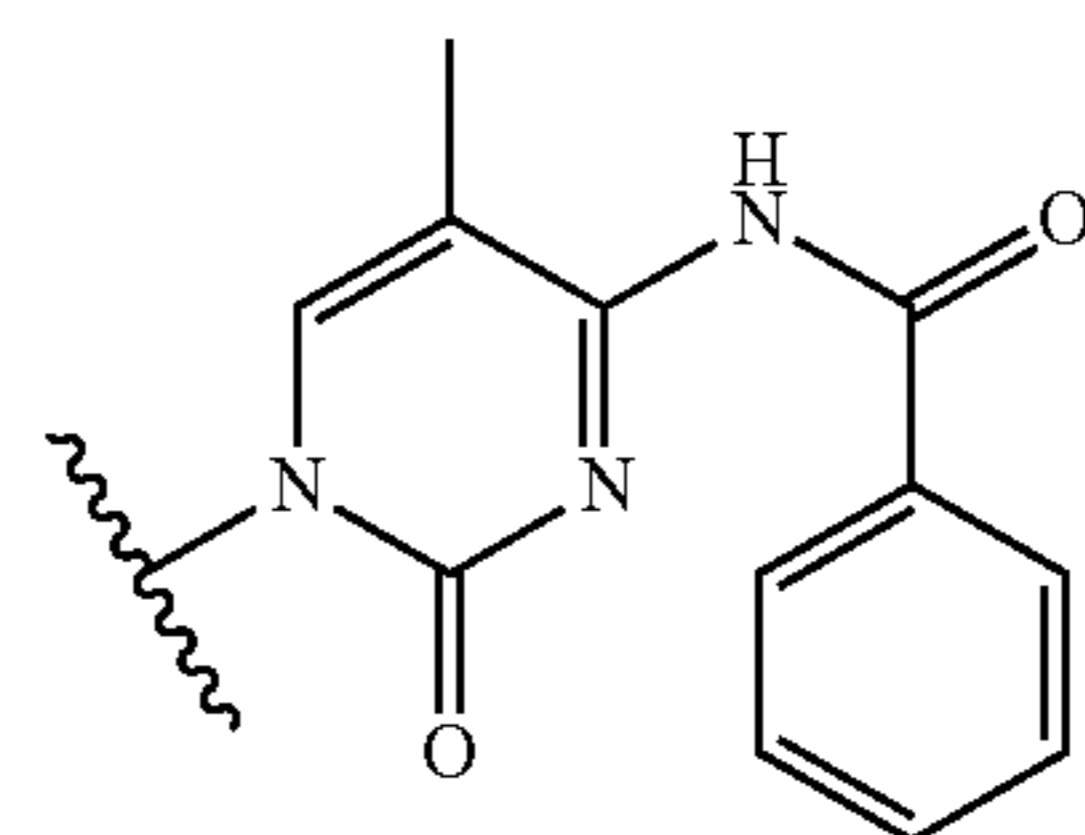
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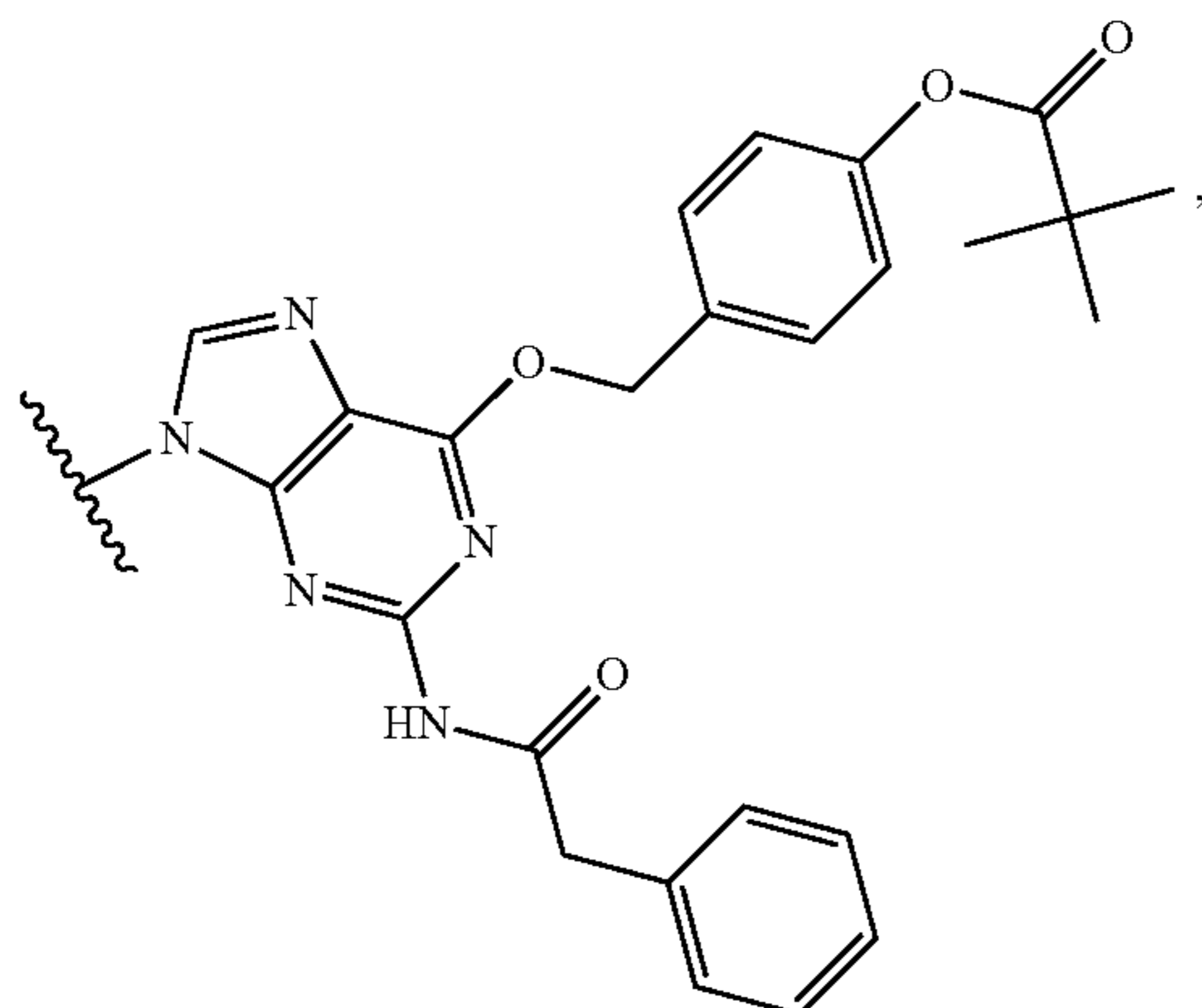
(PC)



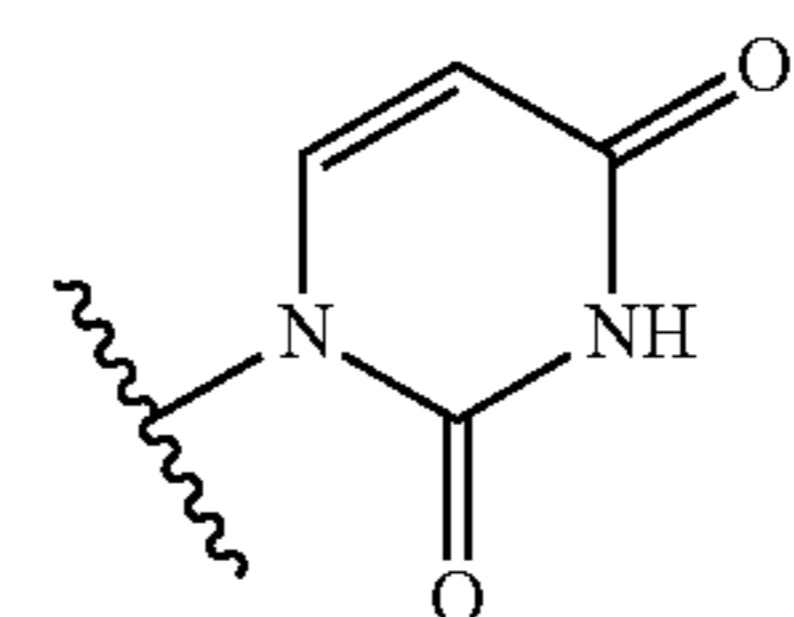
(P5mC)



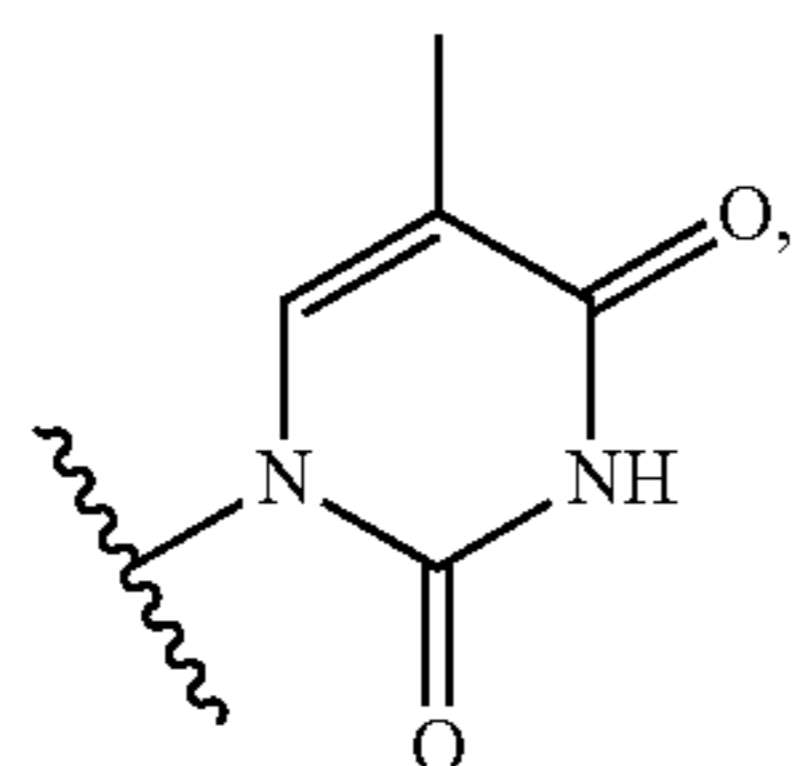
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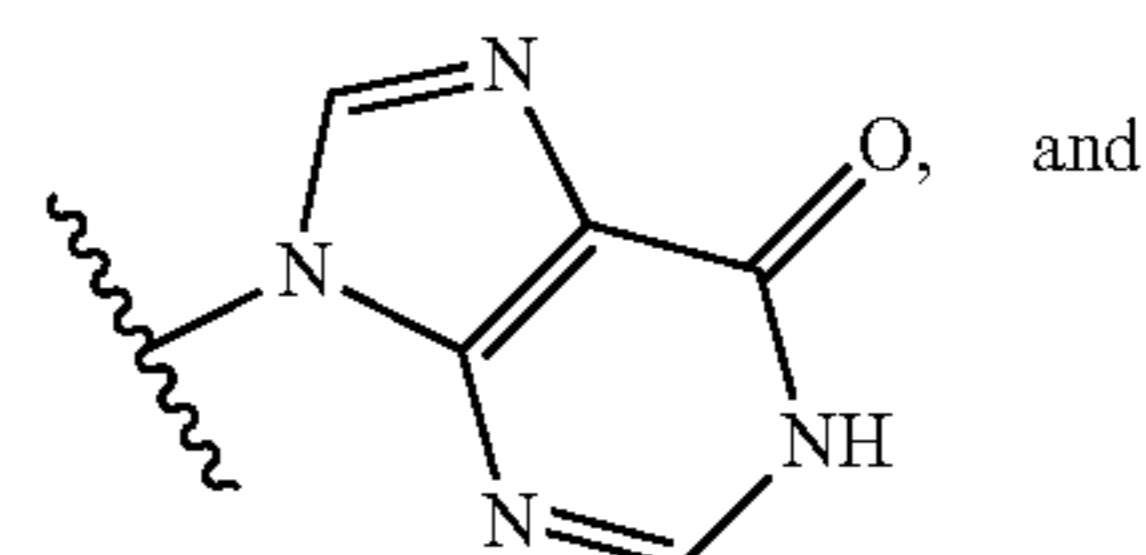
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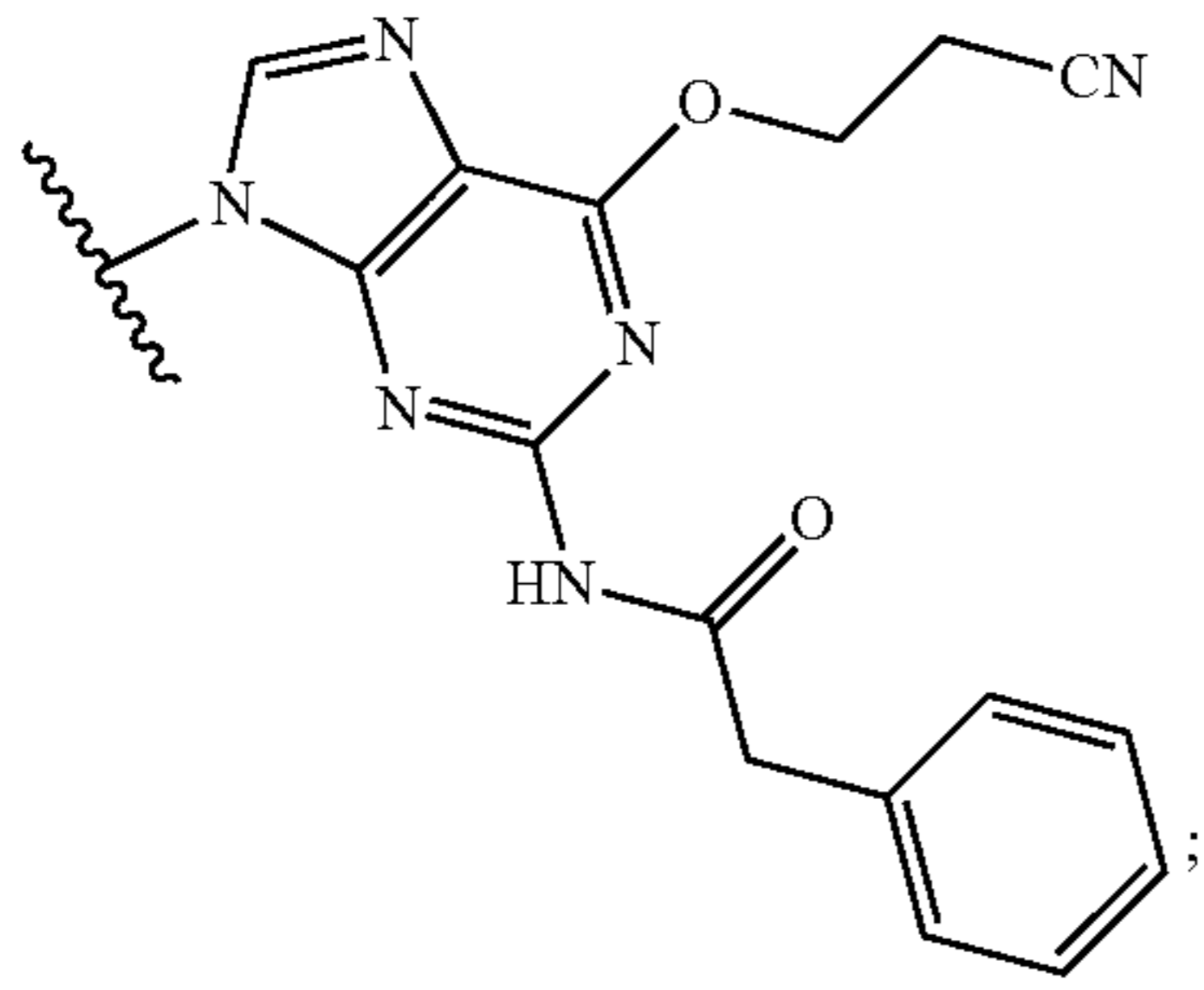


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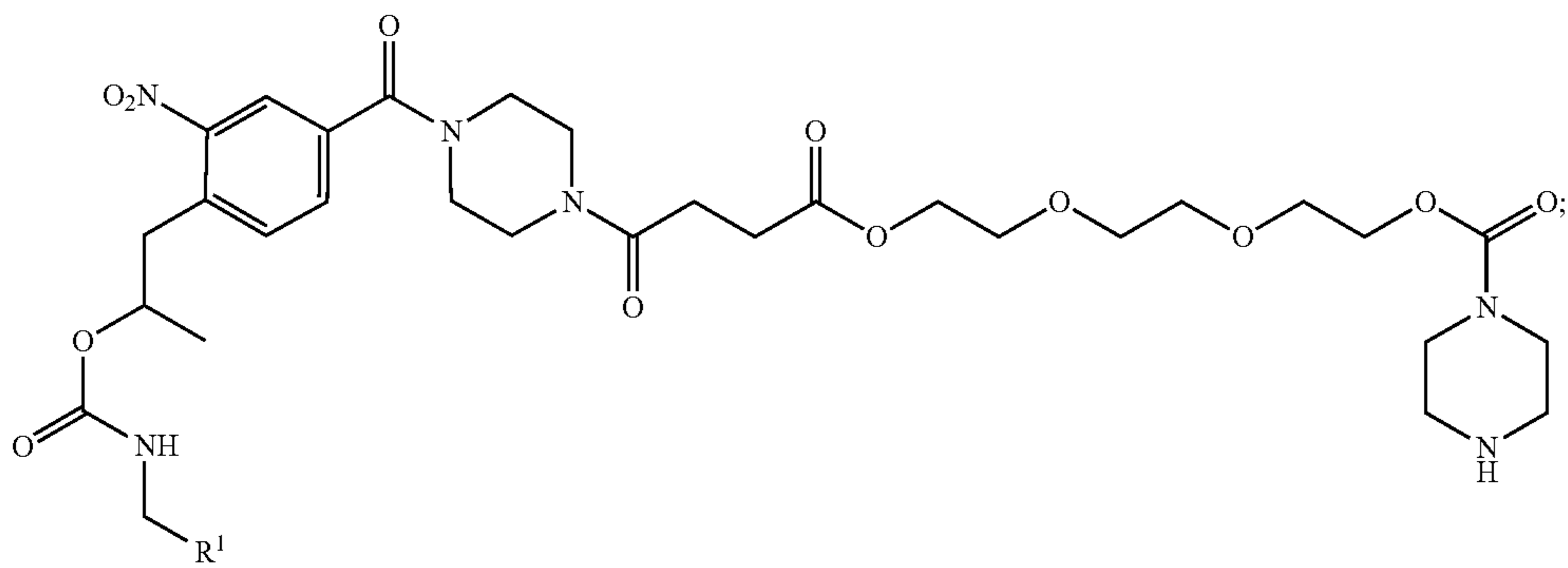
(PG)

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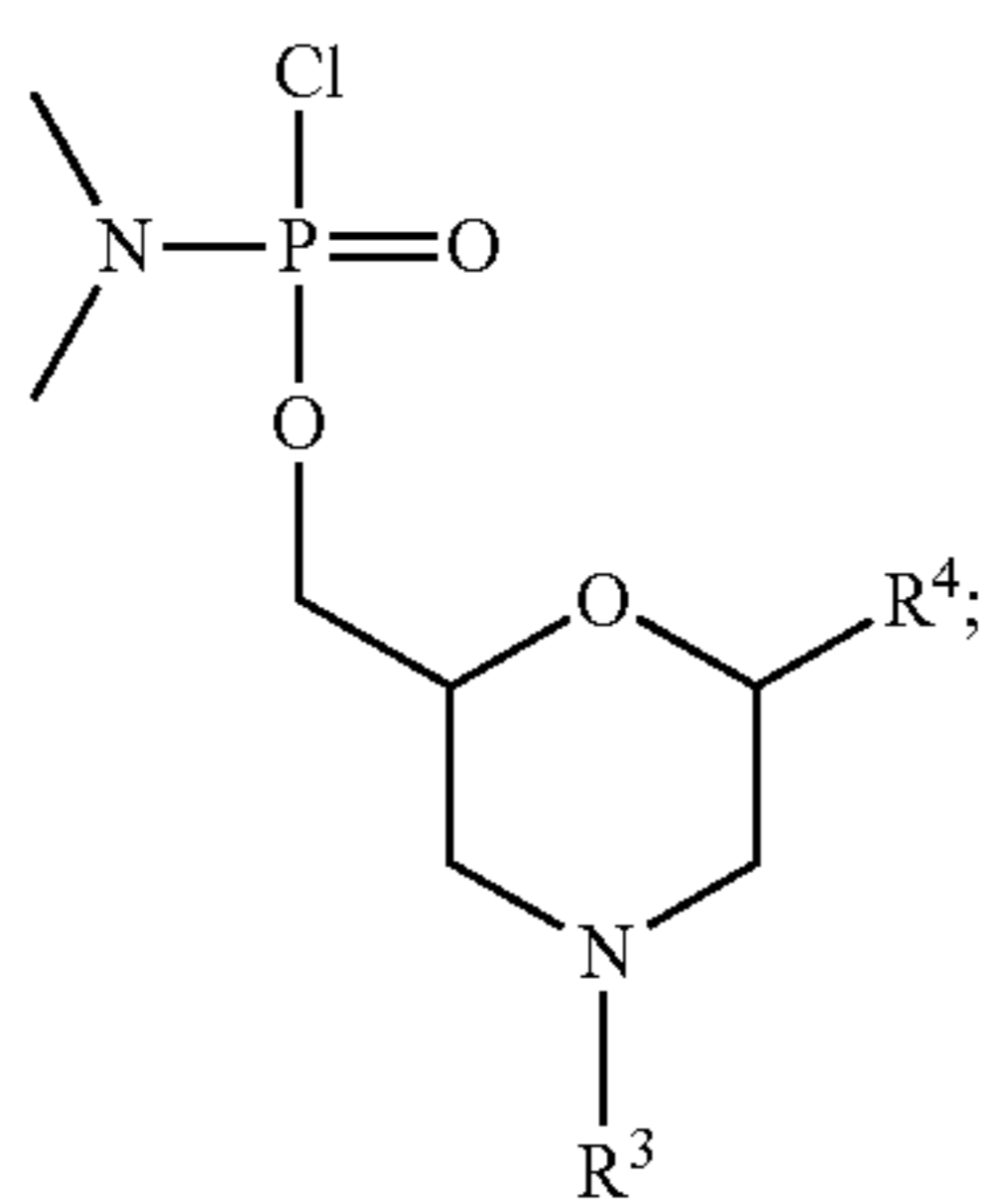
and
wherein the process comprises the sequential steps of:
(a) contacting a compound of Formula (IV):



(IV)

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wherein R¹ is a support-medium;
with a compound of Formula (A4):



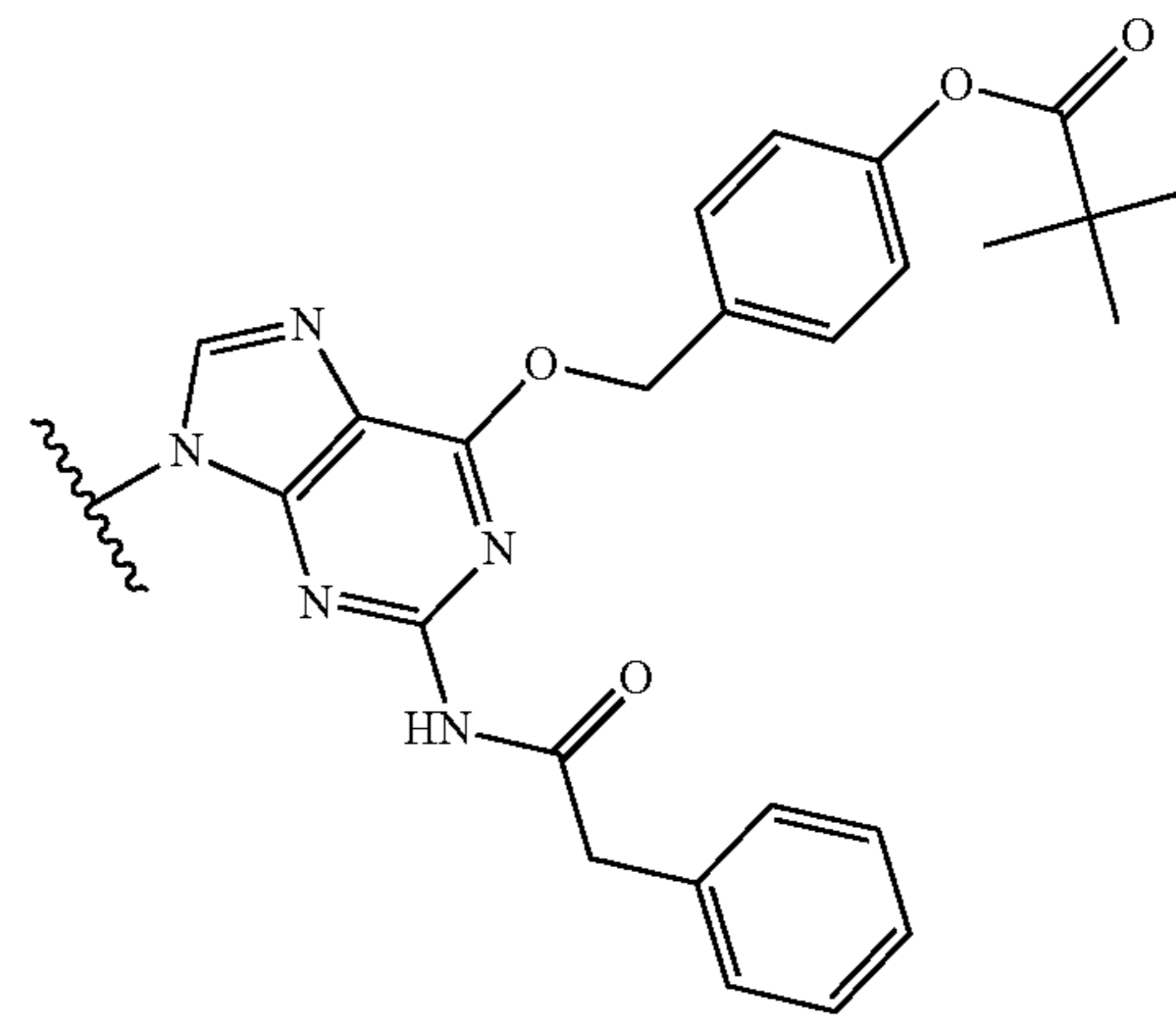
(A4)

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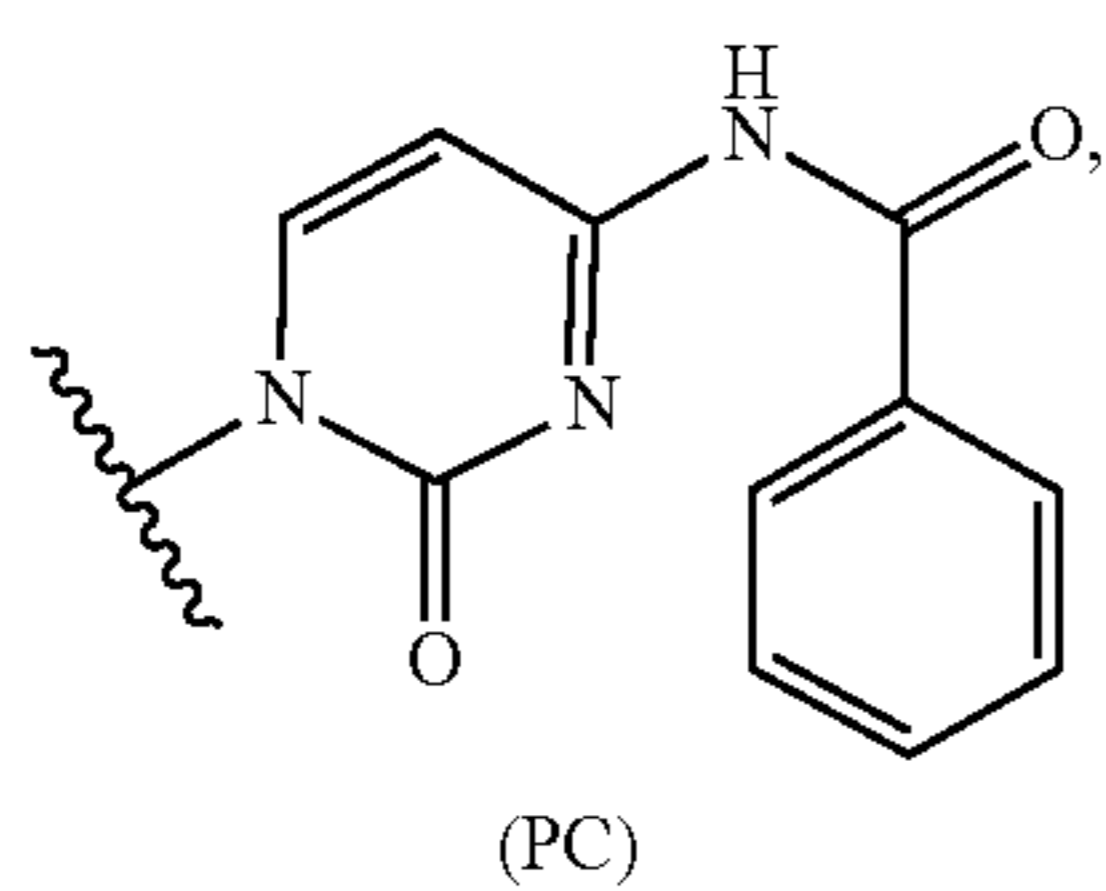
50

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(DPG)

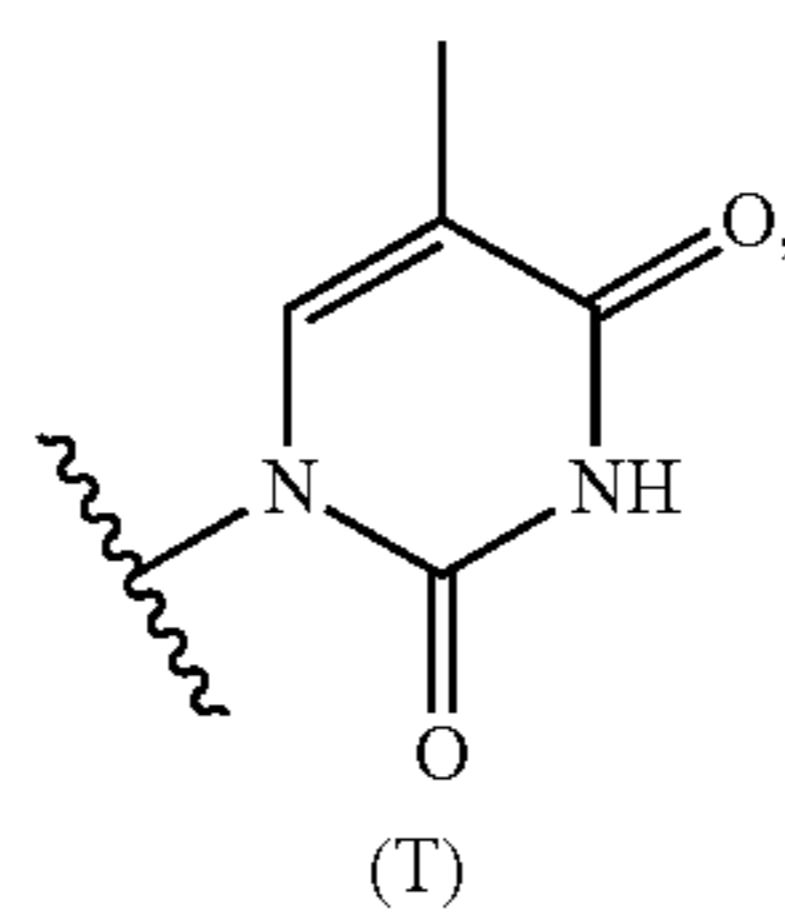
wherein R³ is selected from the group consisting of trityl,
monomethoxytrityl, dimethoxytrityl and trimethoxytrityl,
and R⁴ is selected from the group consisting of:



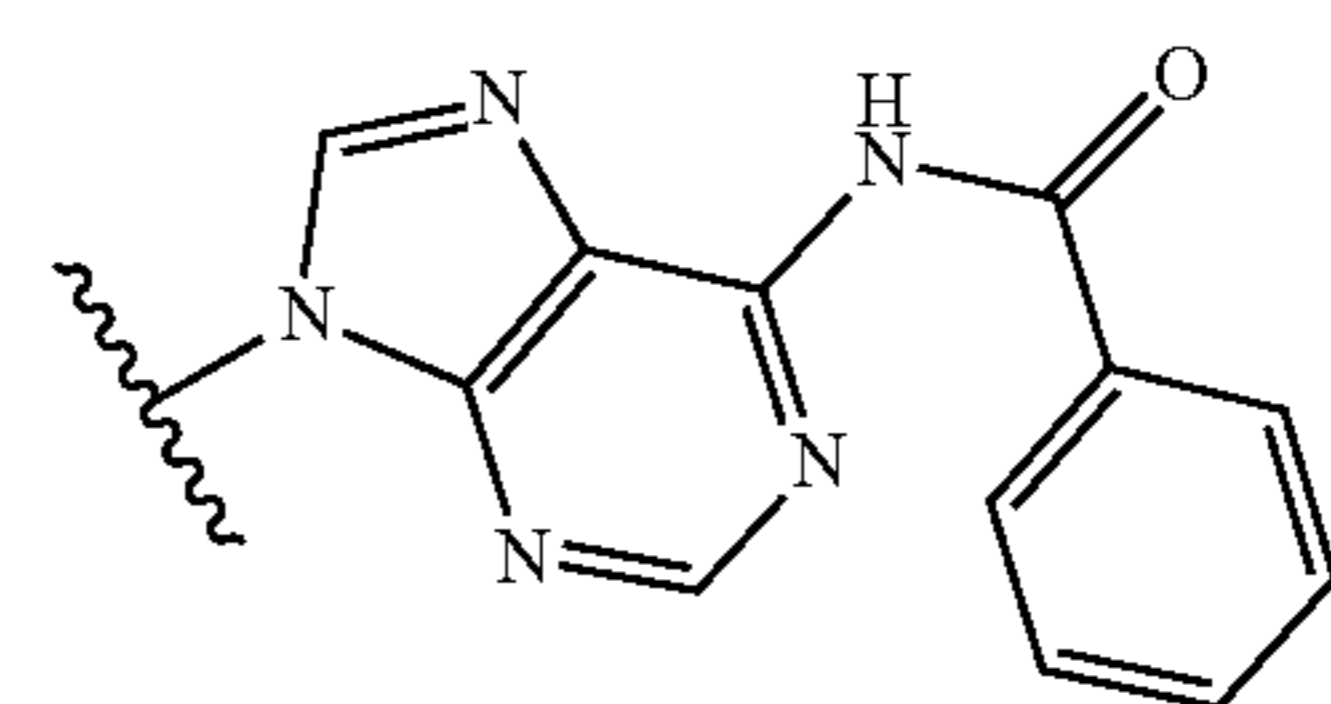
(PC)

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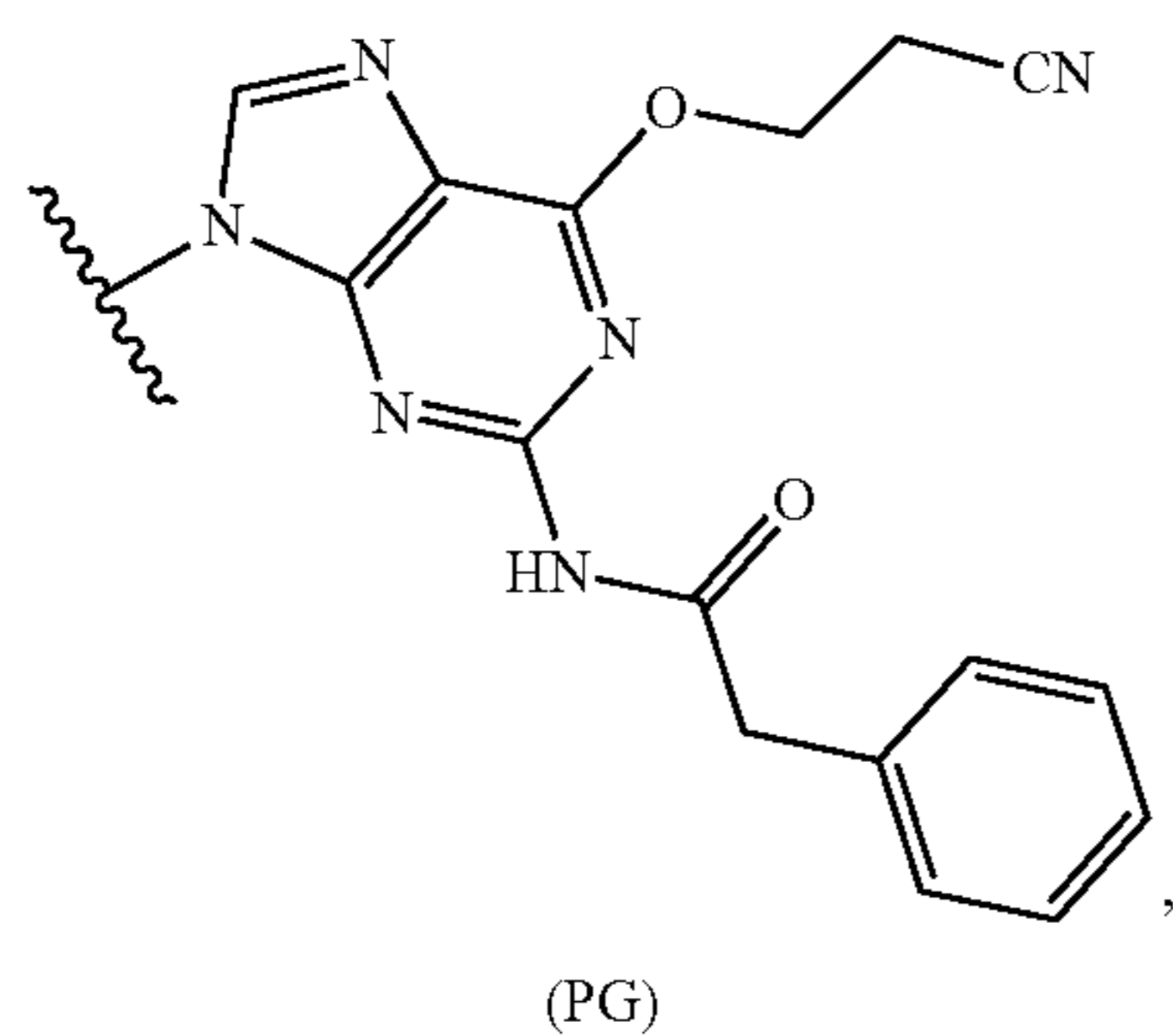
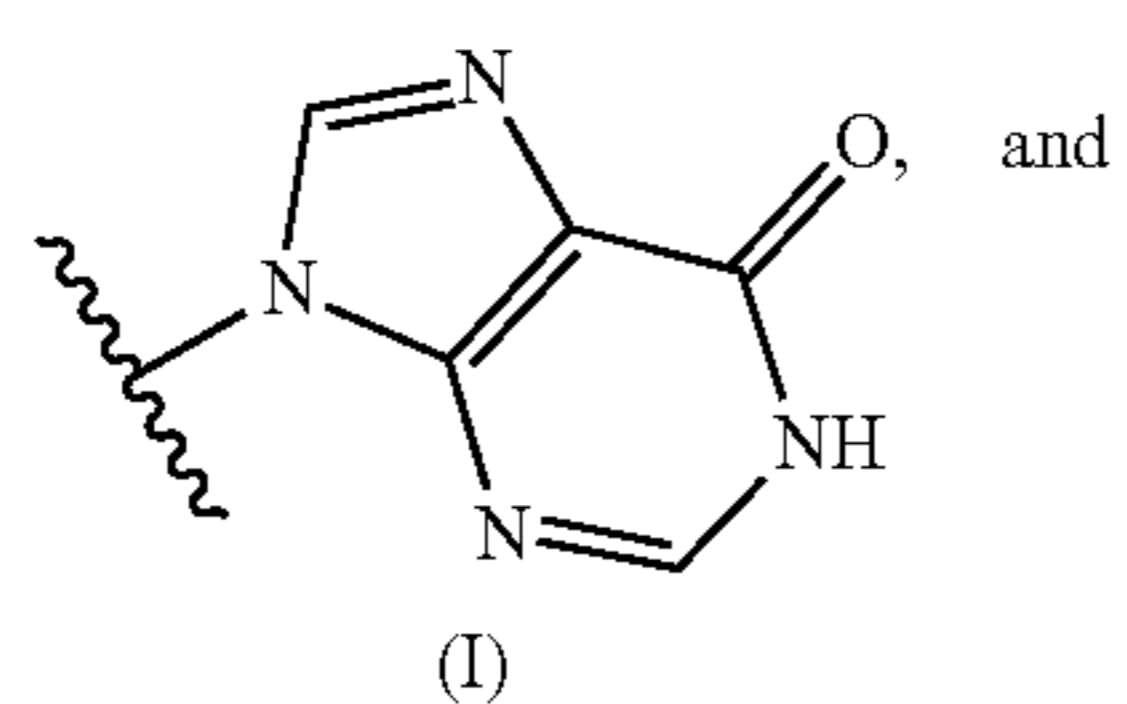
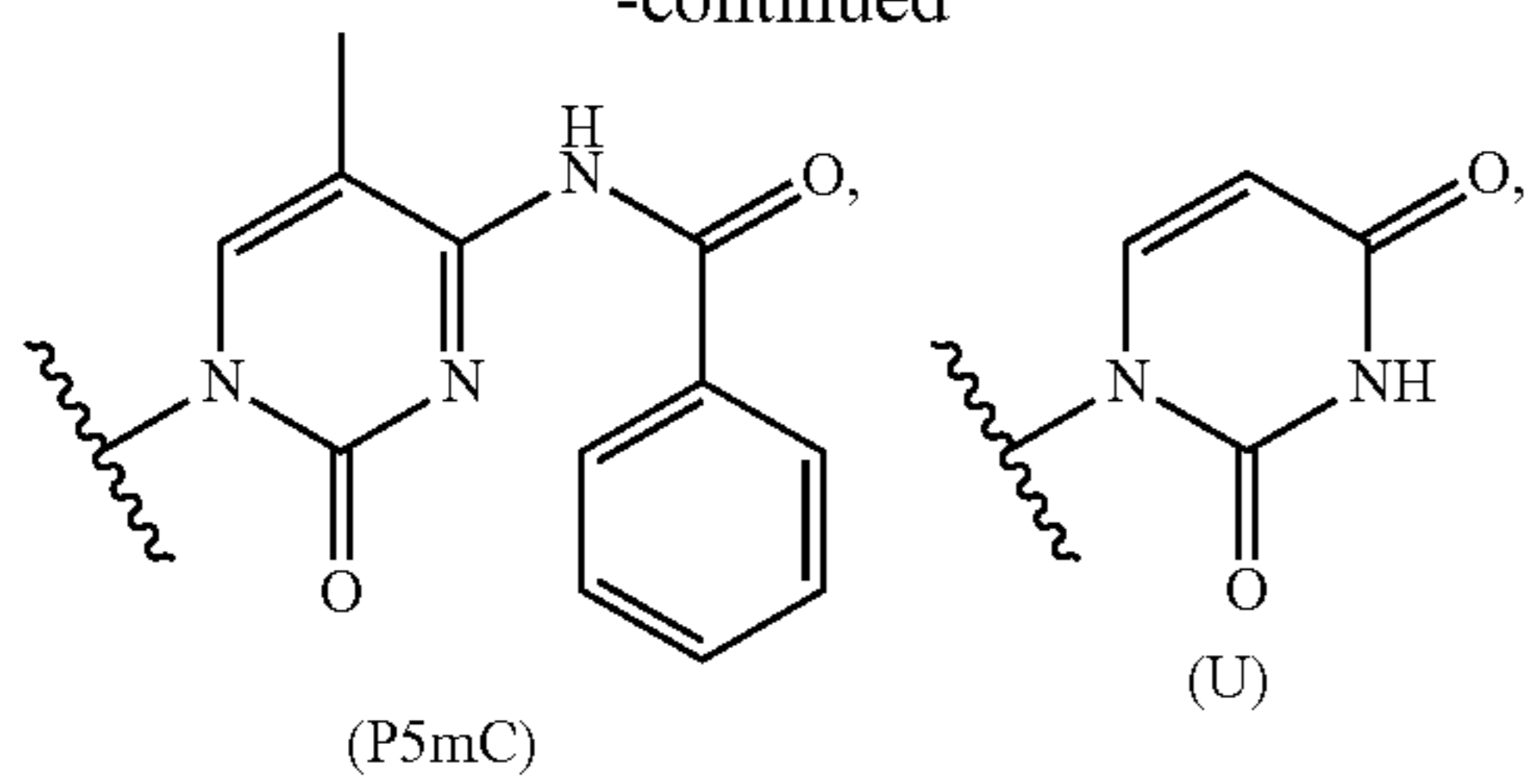
(T)



(PA)

31

-continued



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wherein R^1 is a support-medium, R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

R^4 is selected from the group consisting of:

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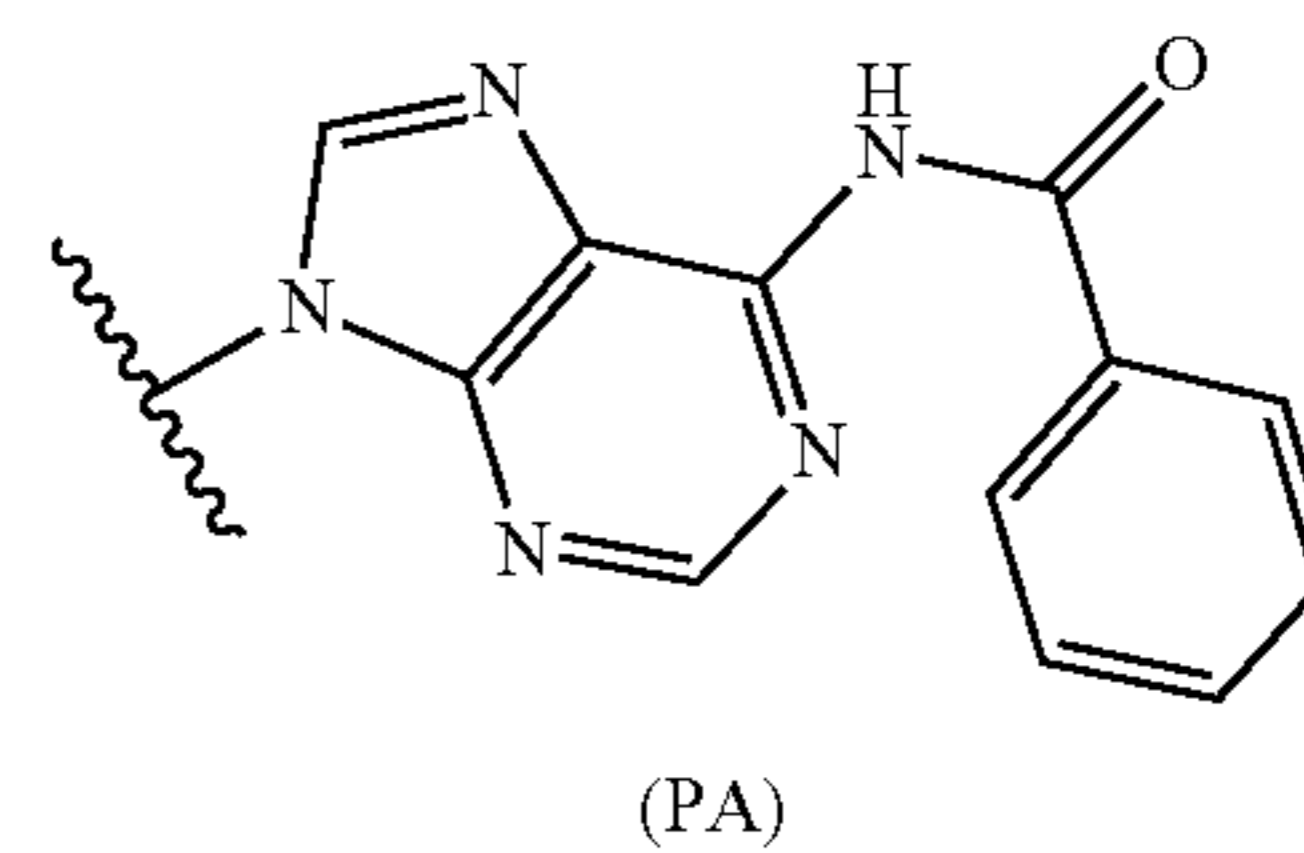
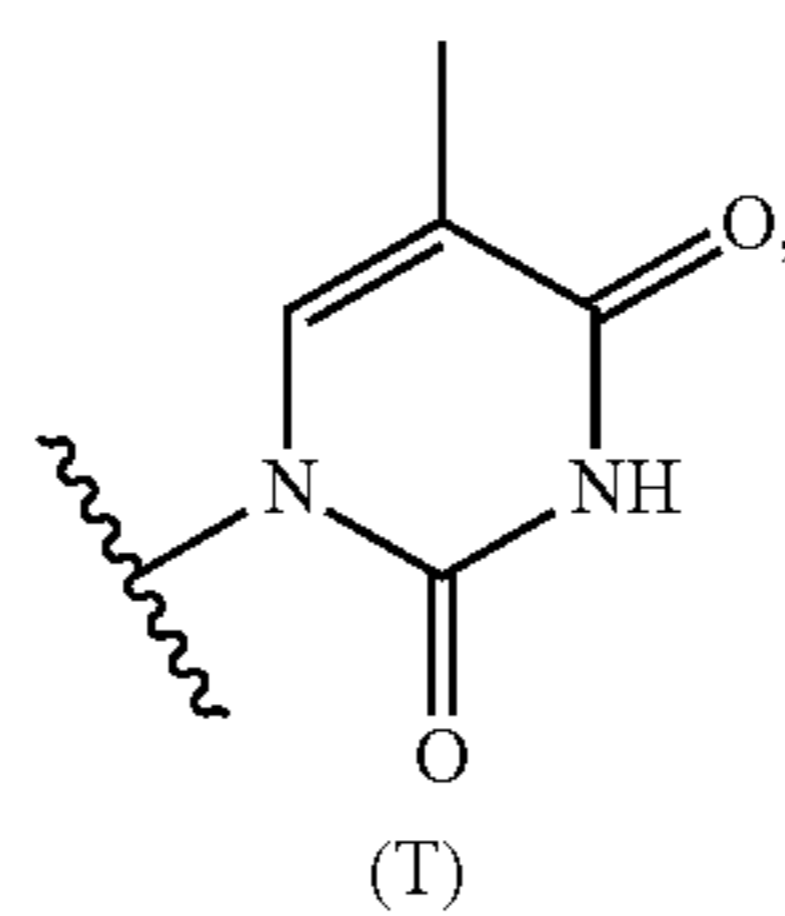
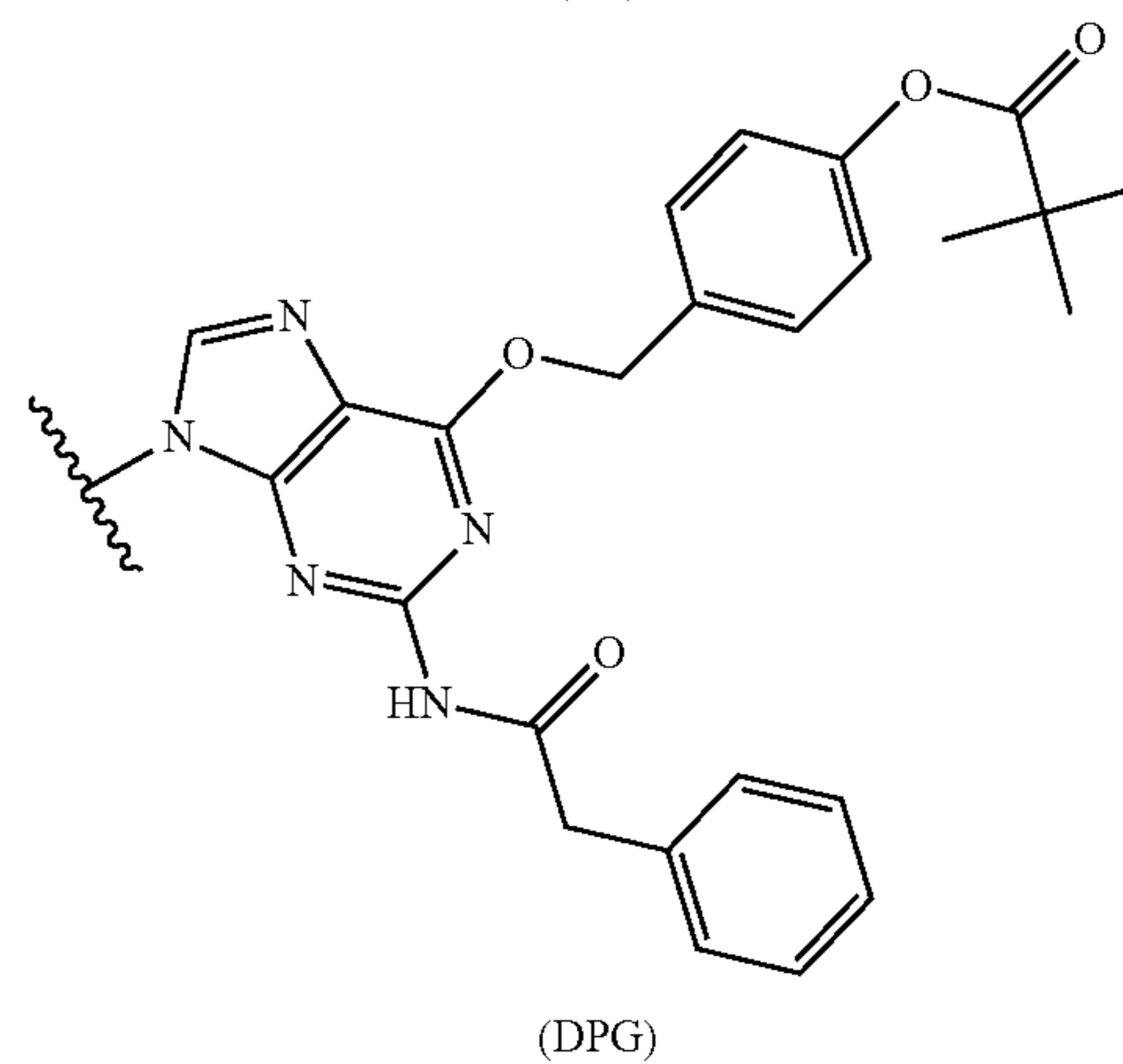
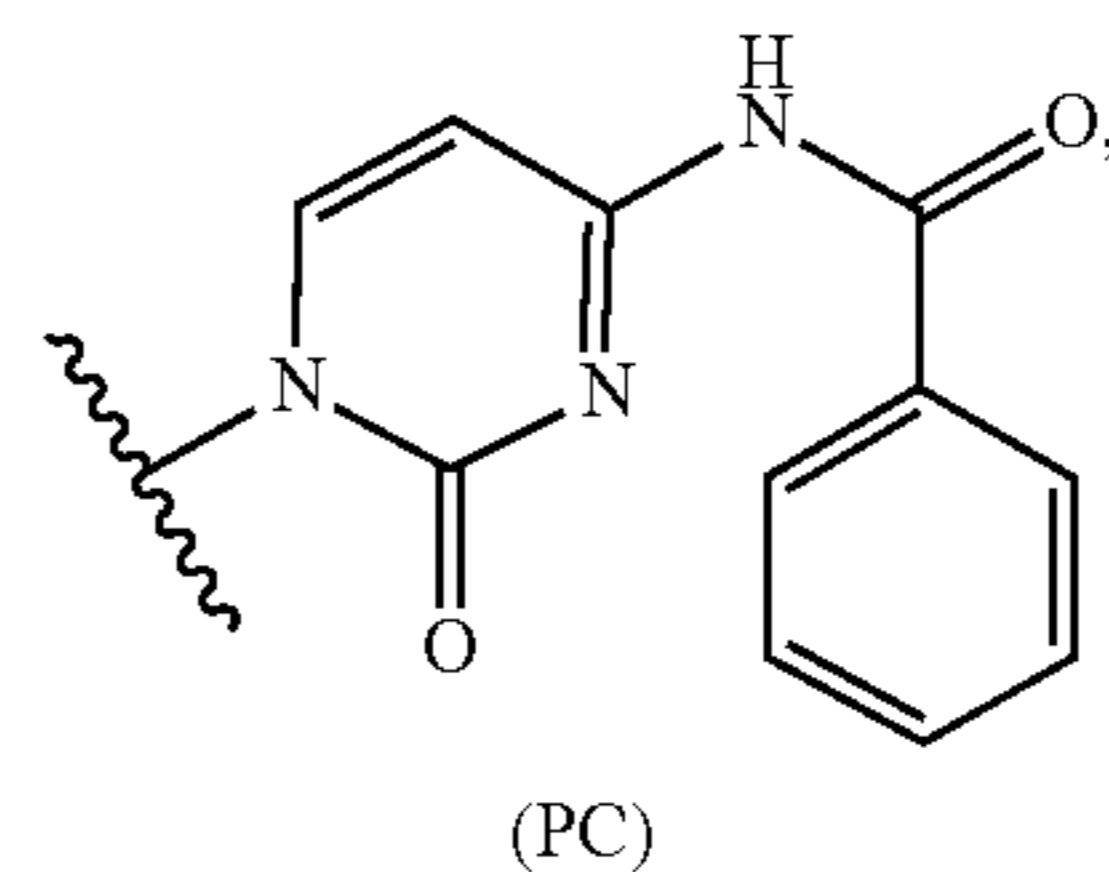
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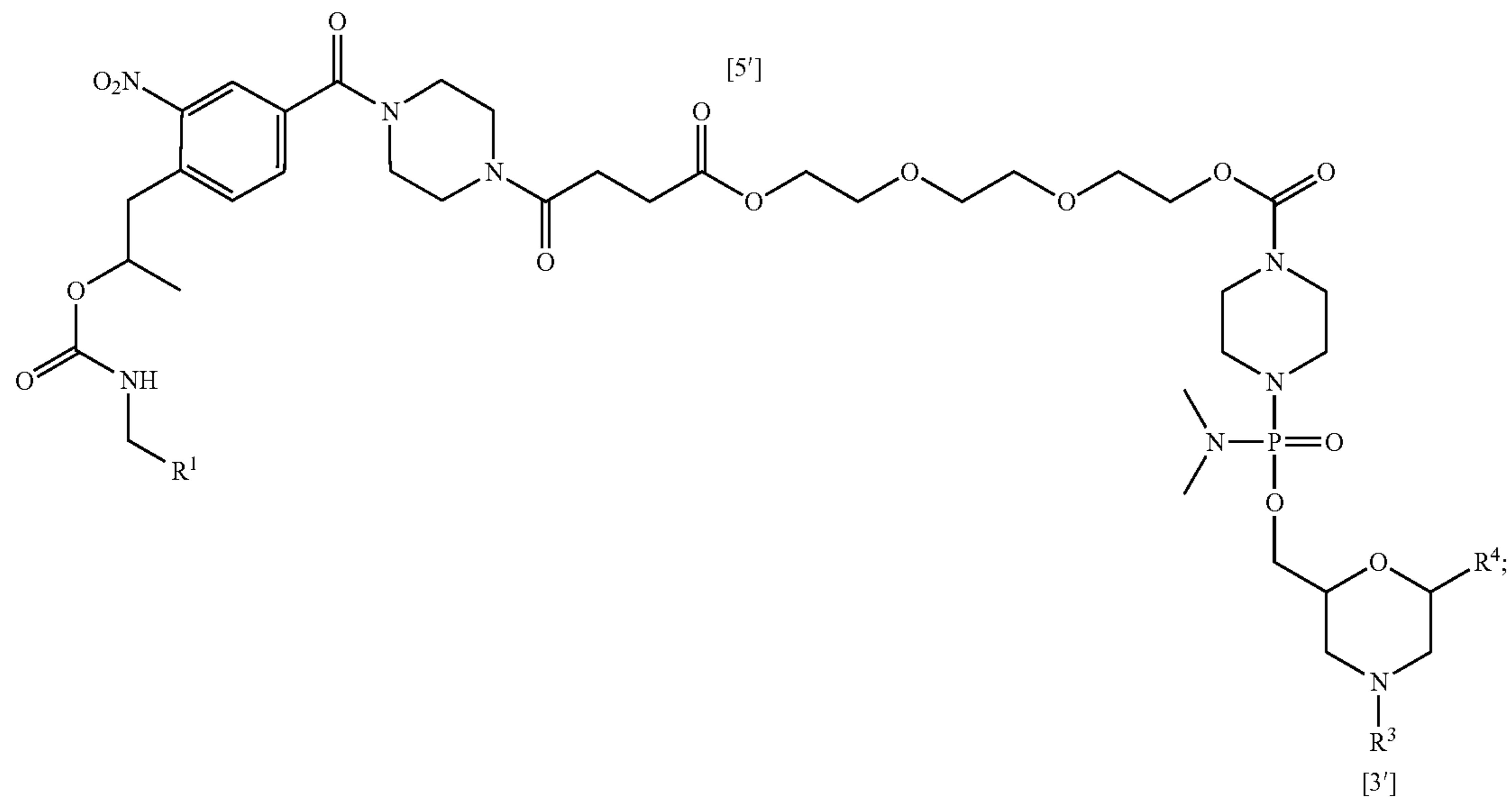
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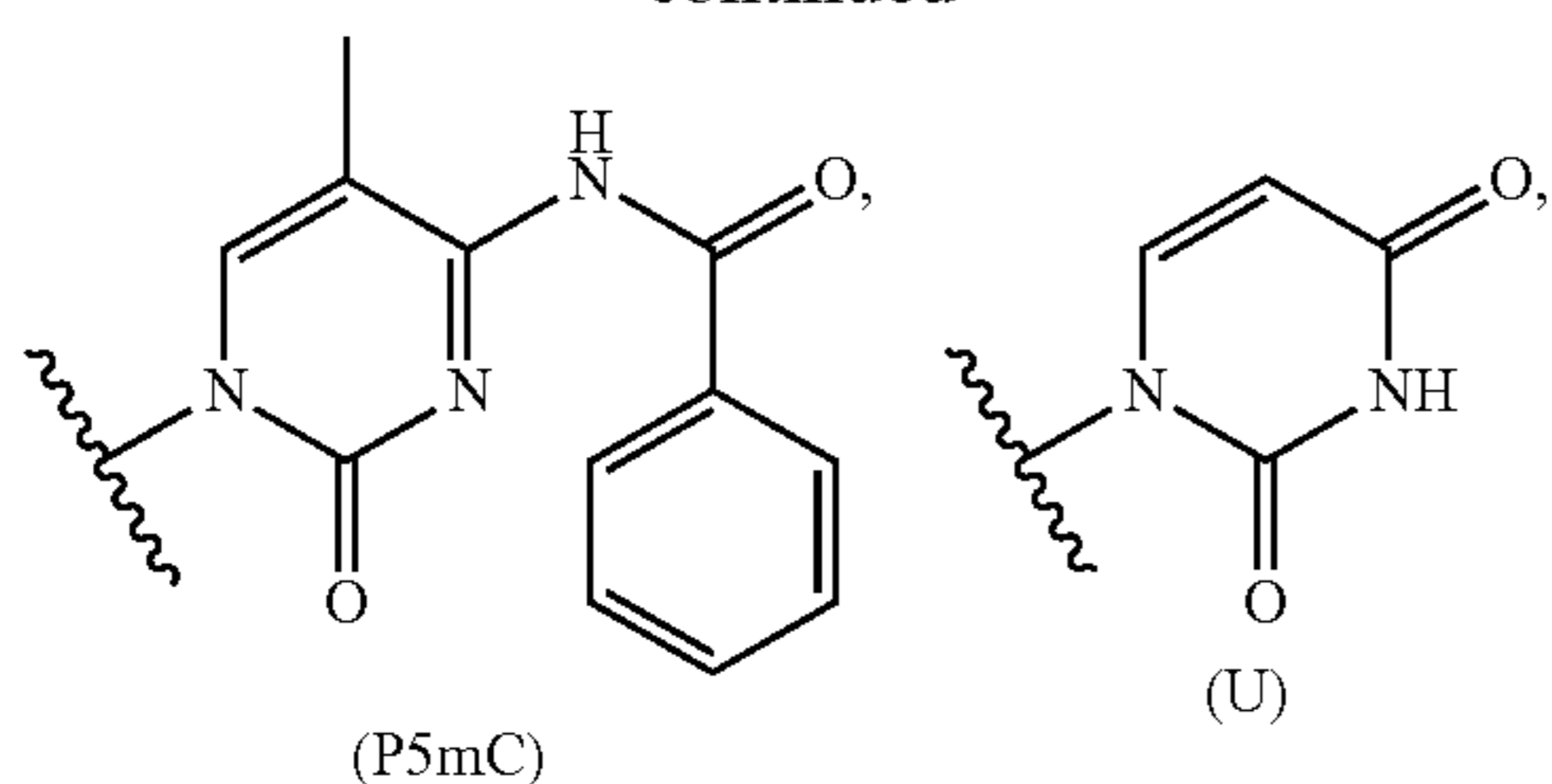


to form a compound of Formula (A5):



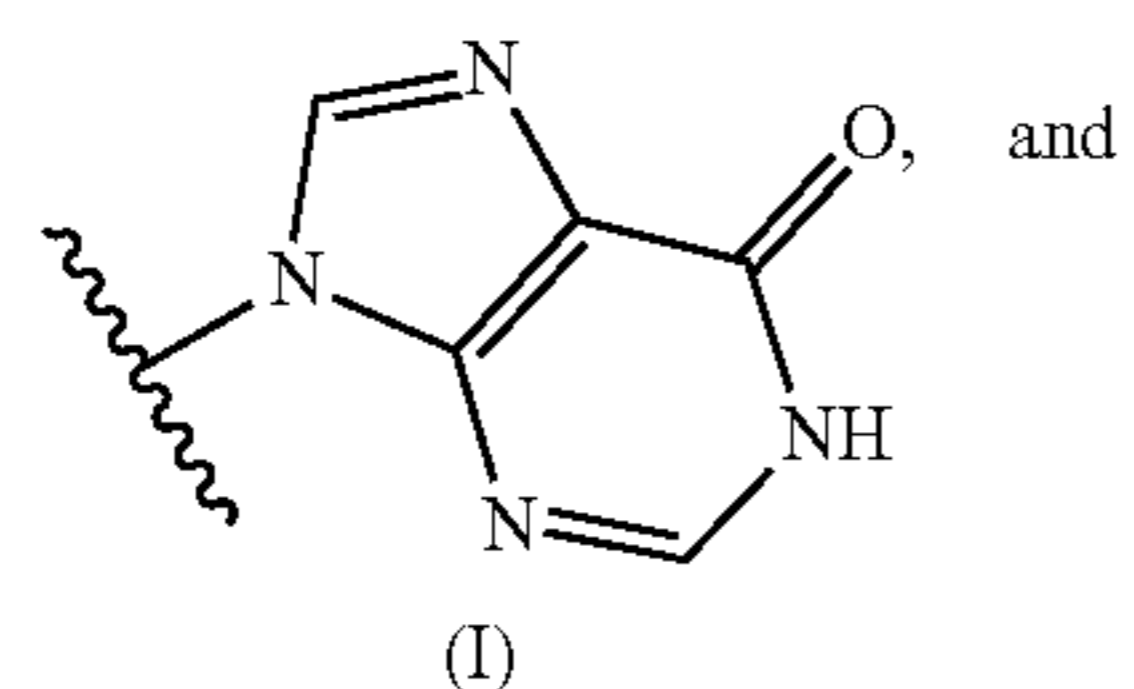
33

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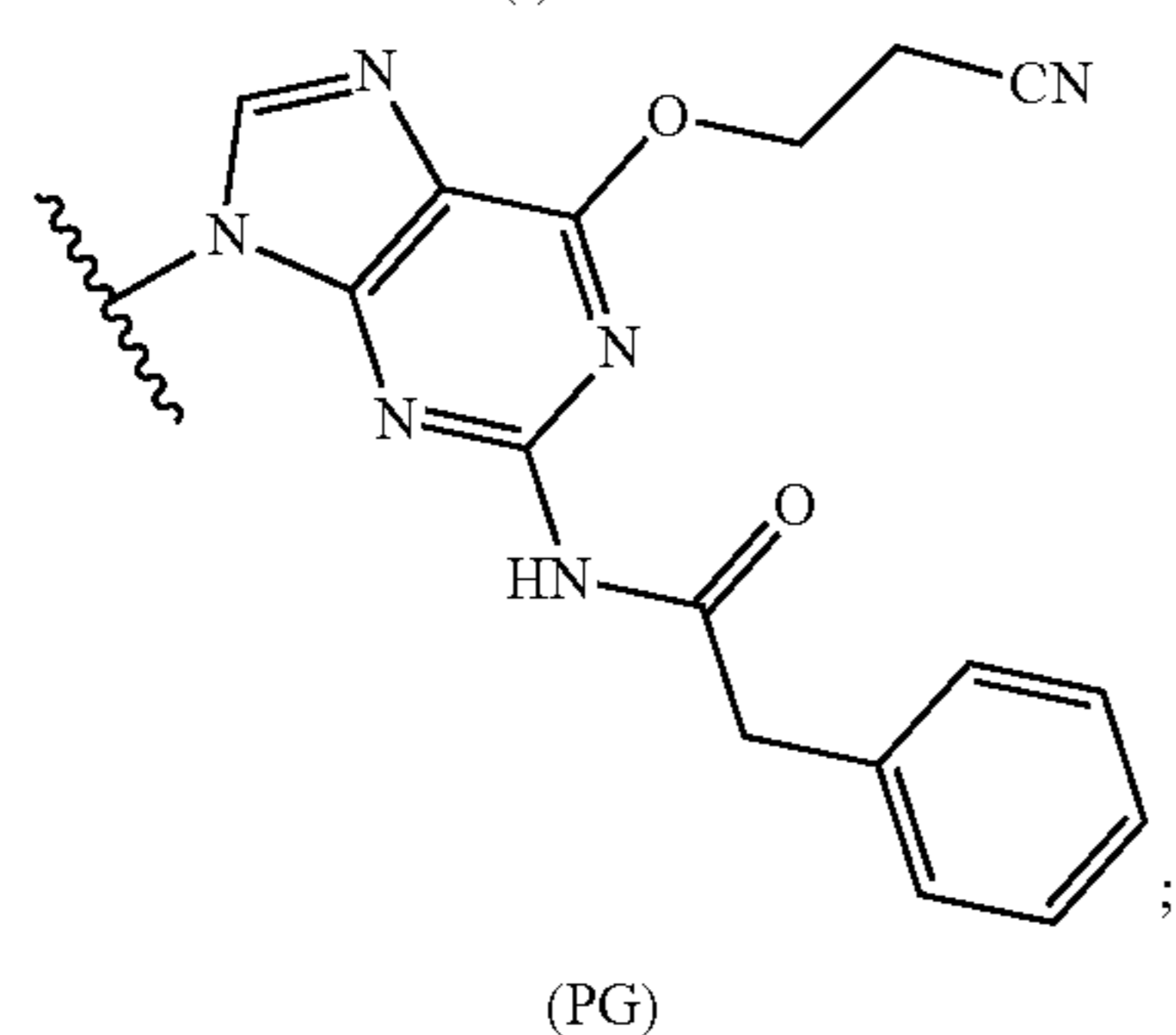


(P5mC)

(U)



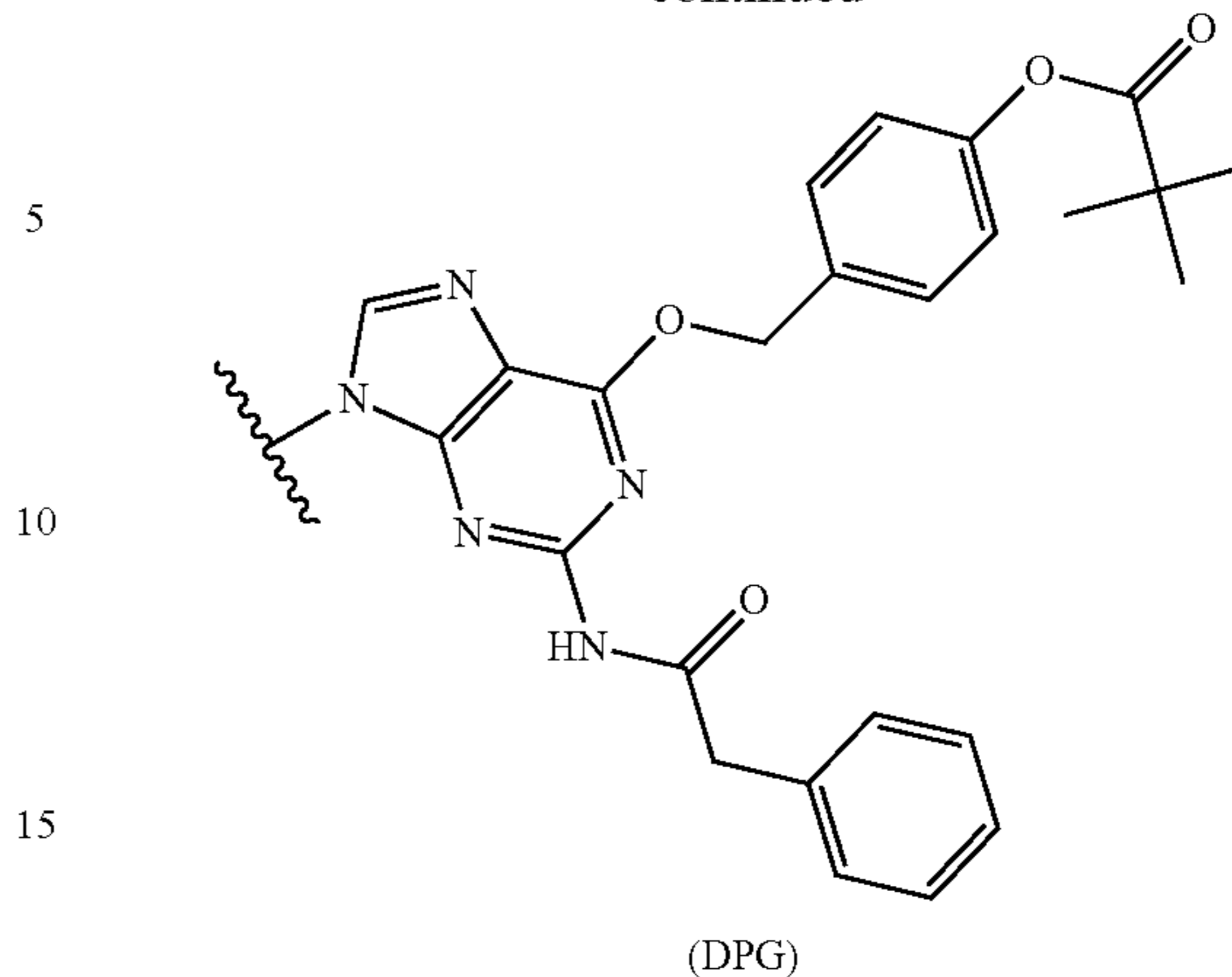
(I)



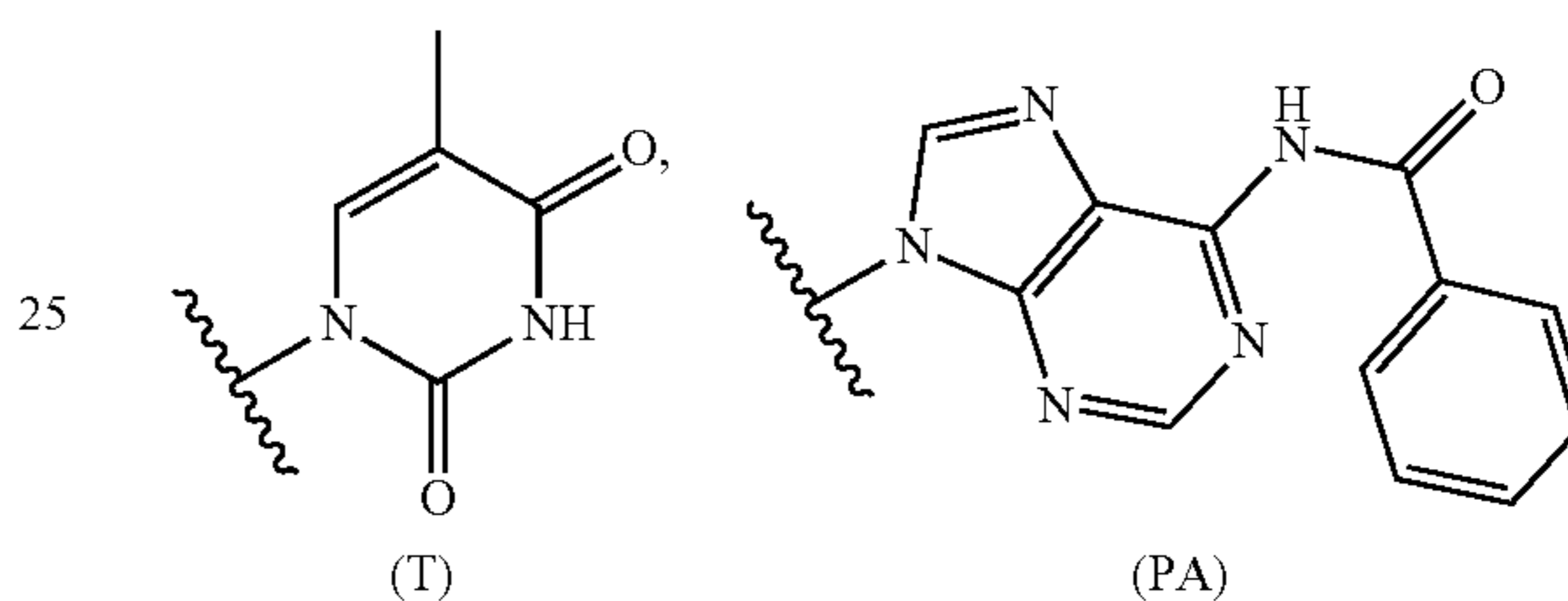
(PG)

34

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(DPG)



(T)

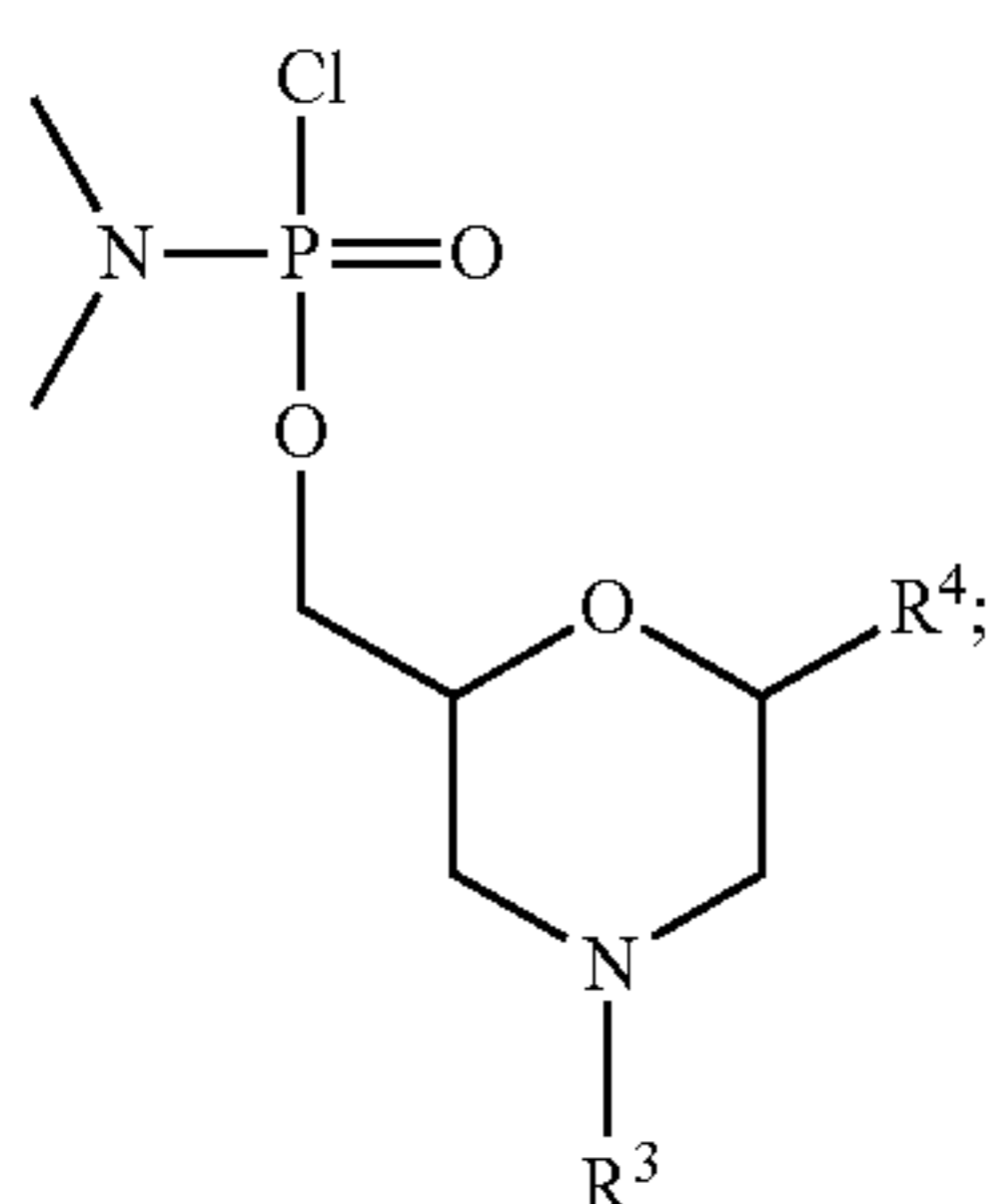
(PA)

and

(b) performing n-1 iterations of the sequential steps of:

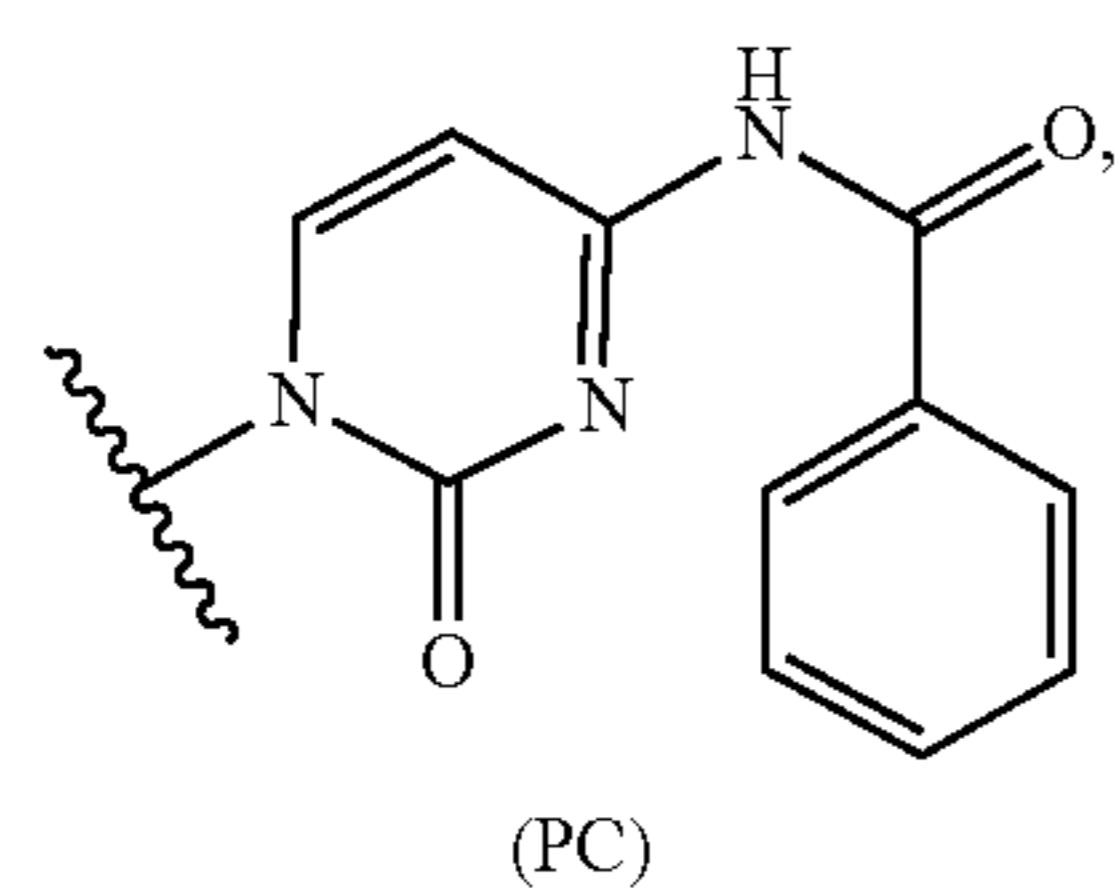
(b1) contacting the product formed by the immediately prior step with a deblocking agent; and

(b2) contacting the compound formed by the immediately prior step with a compound of Formula (A8):

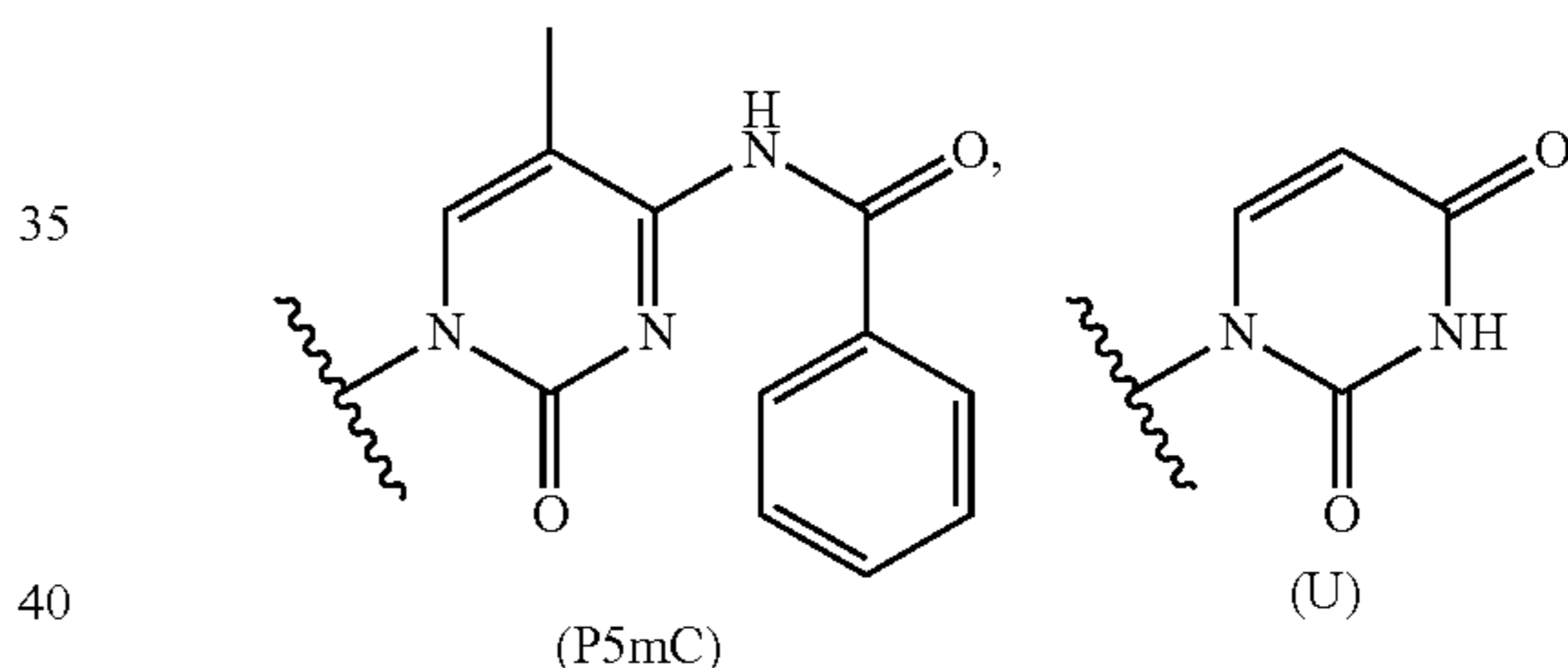


(A8)

wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

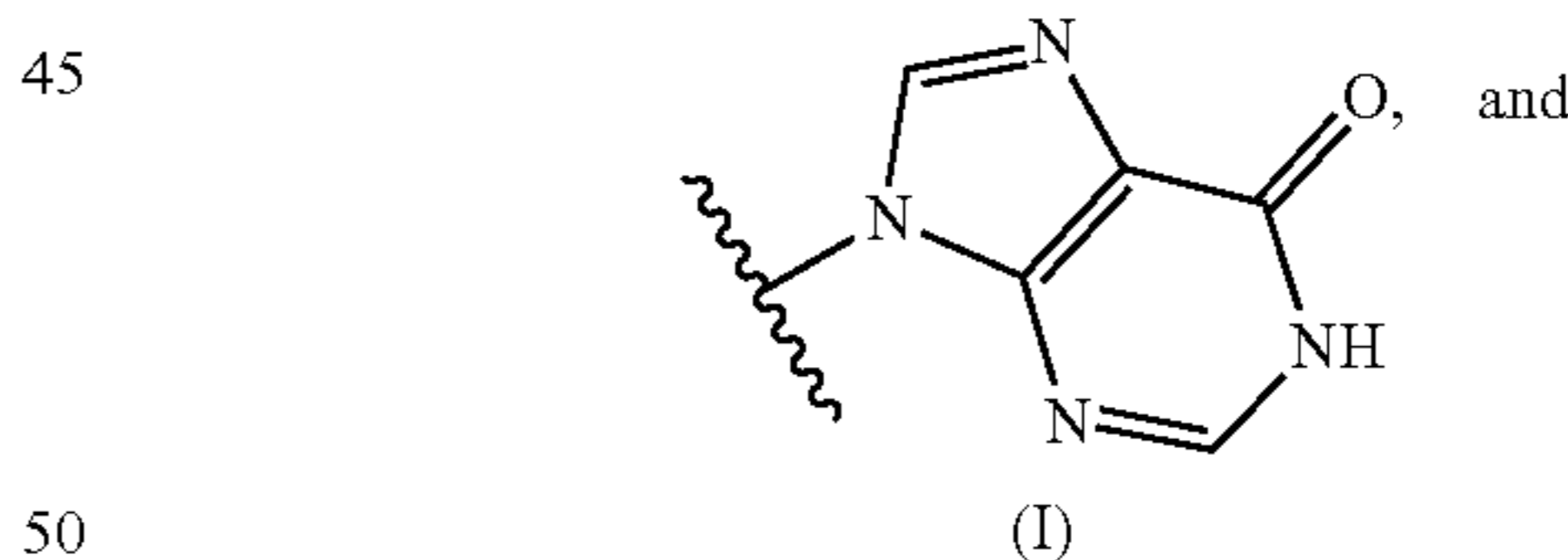


(PC)

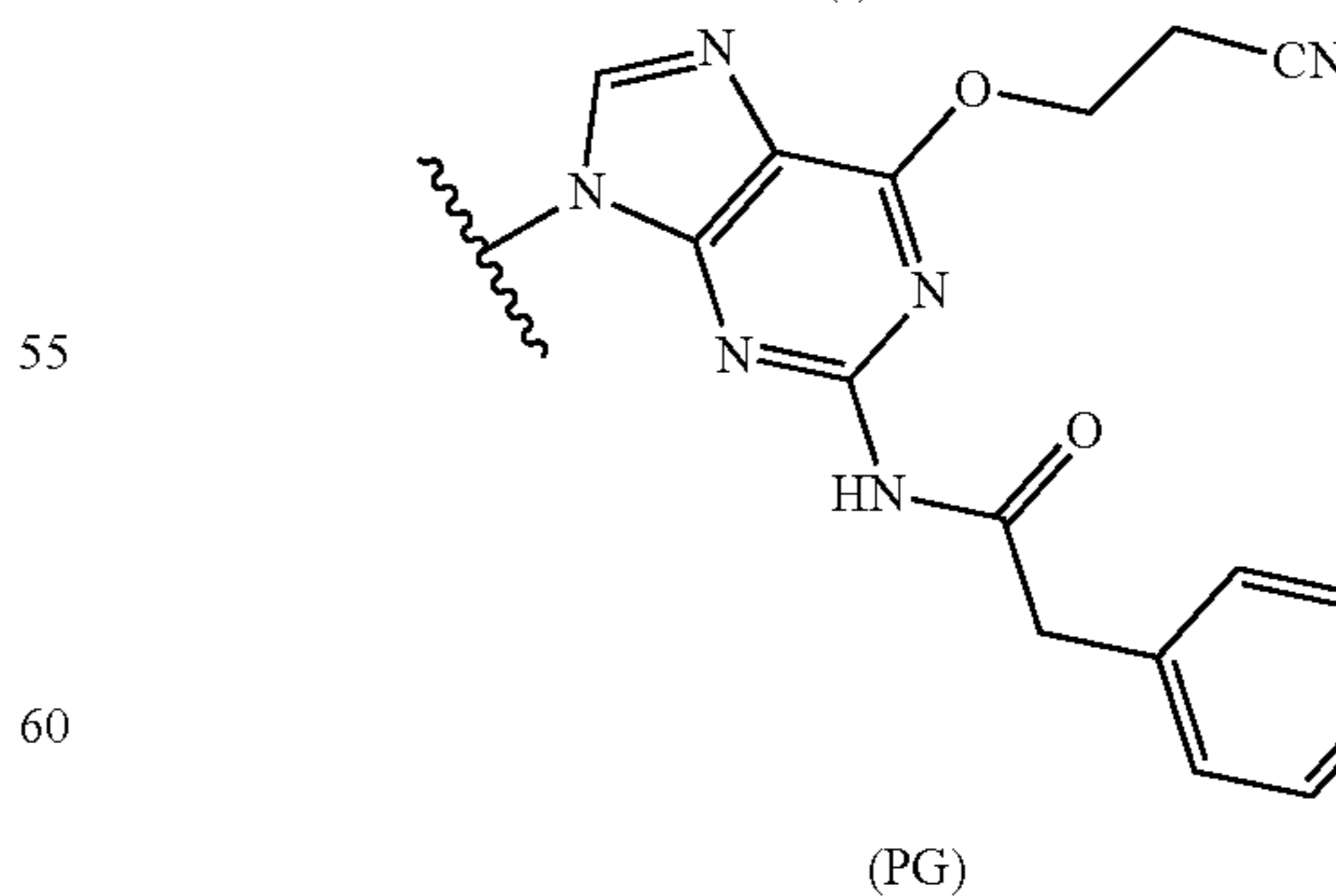


(P5mC)

(U)



(I)



(PG)

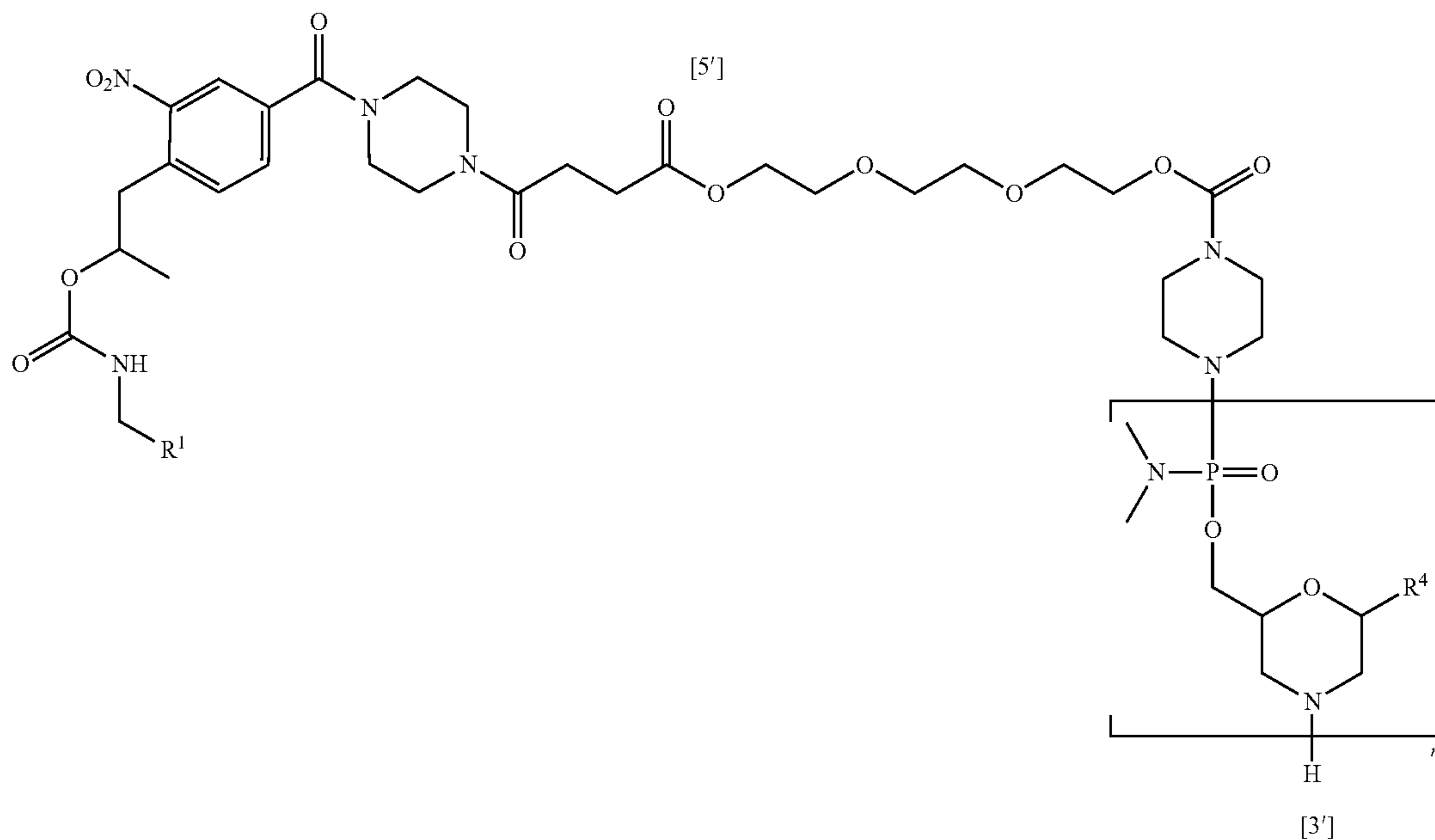
to form a compound of Formula (A9).

In yet another aspect, provided herein is a process for preparing a compound of Formula (A10):

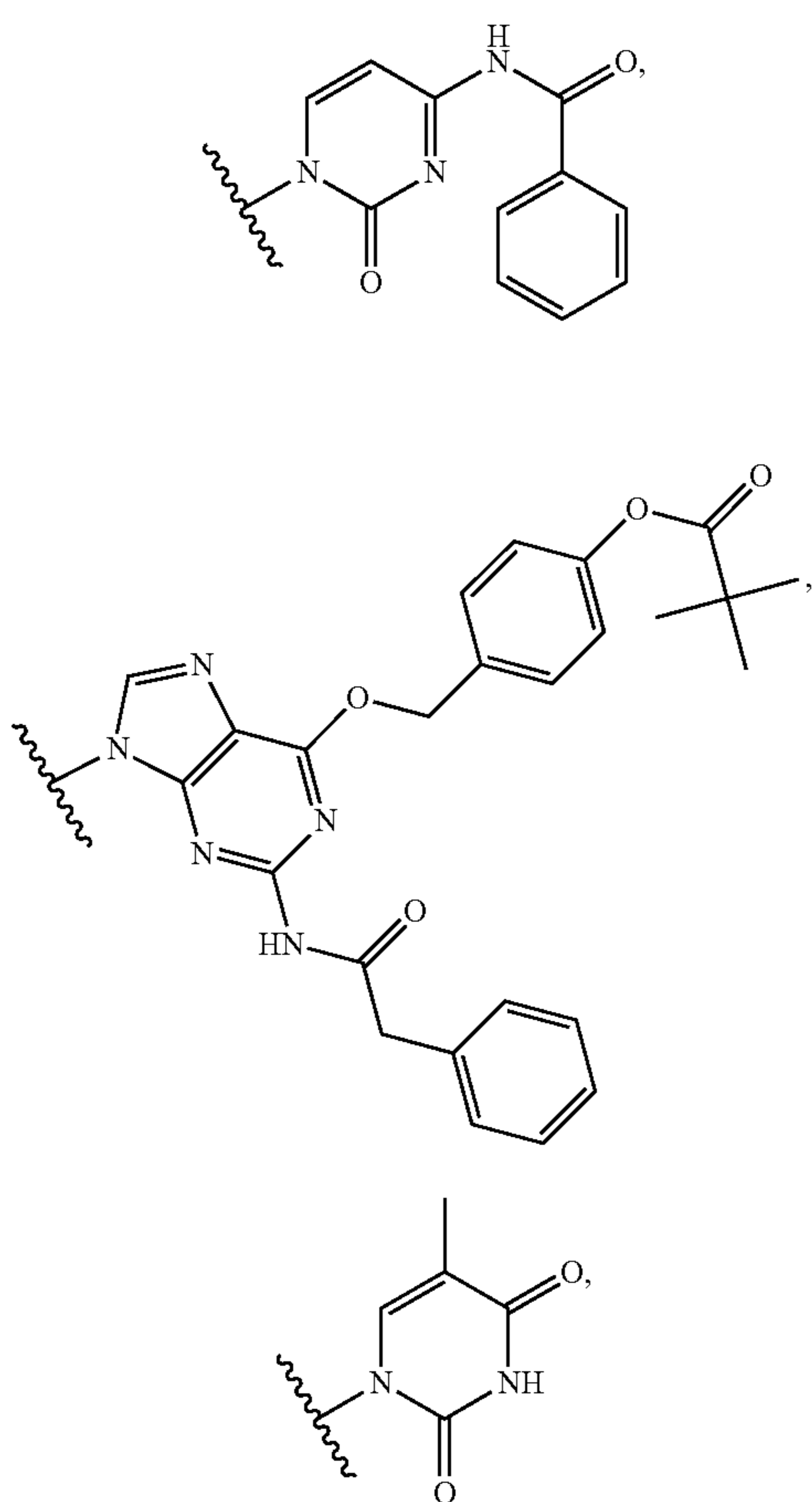
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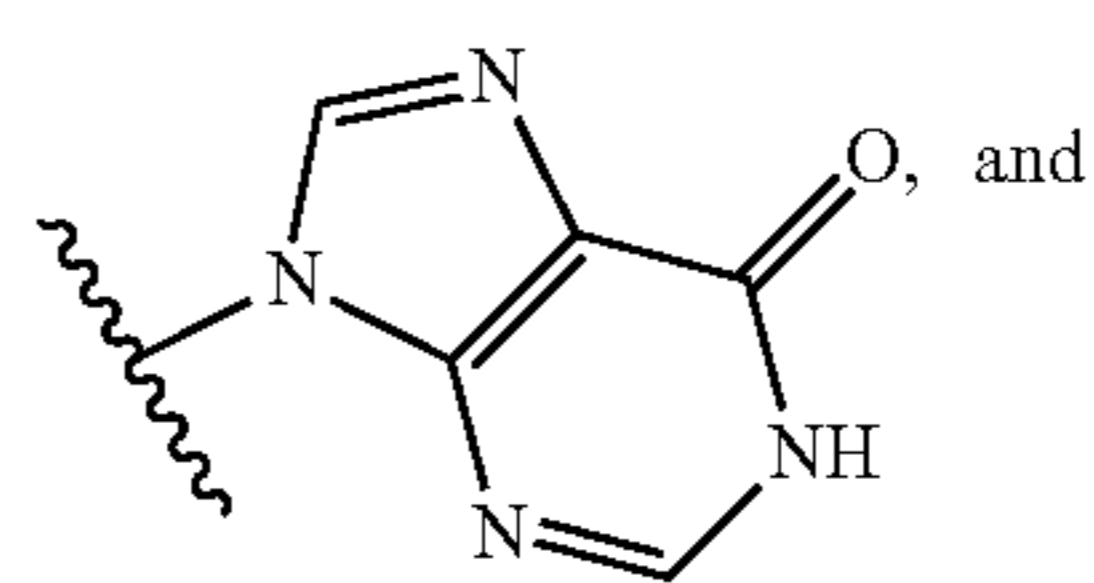
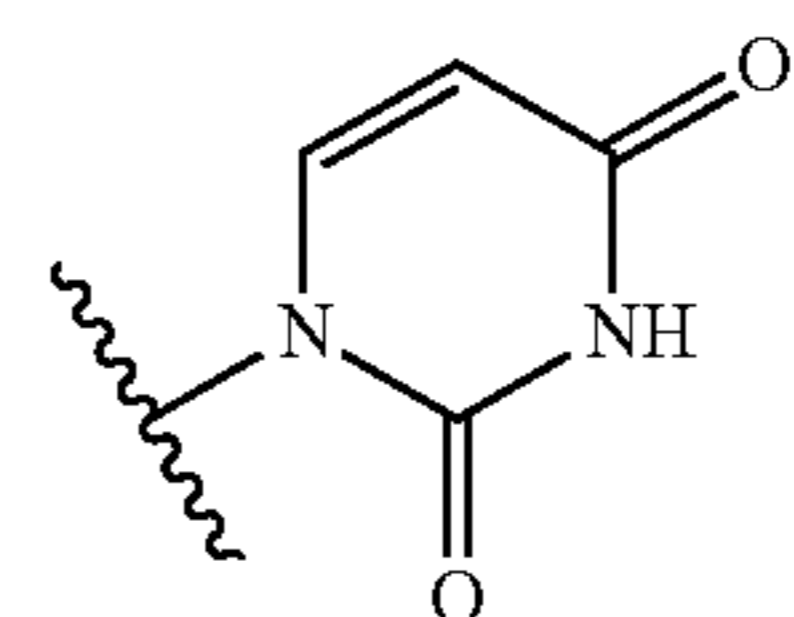
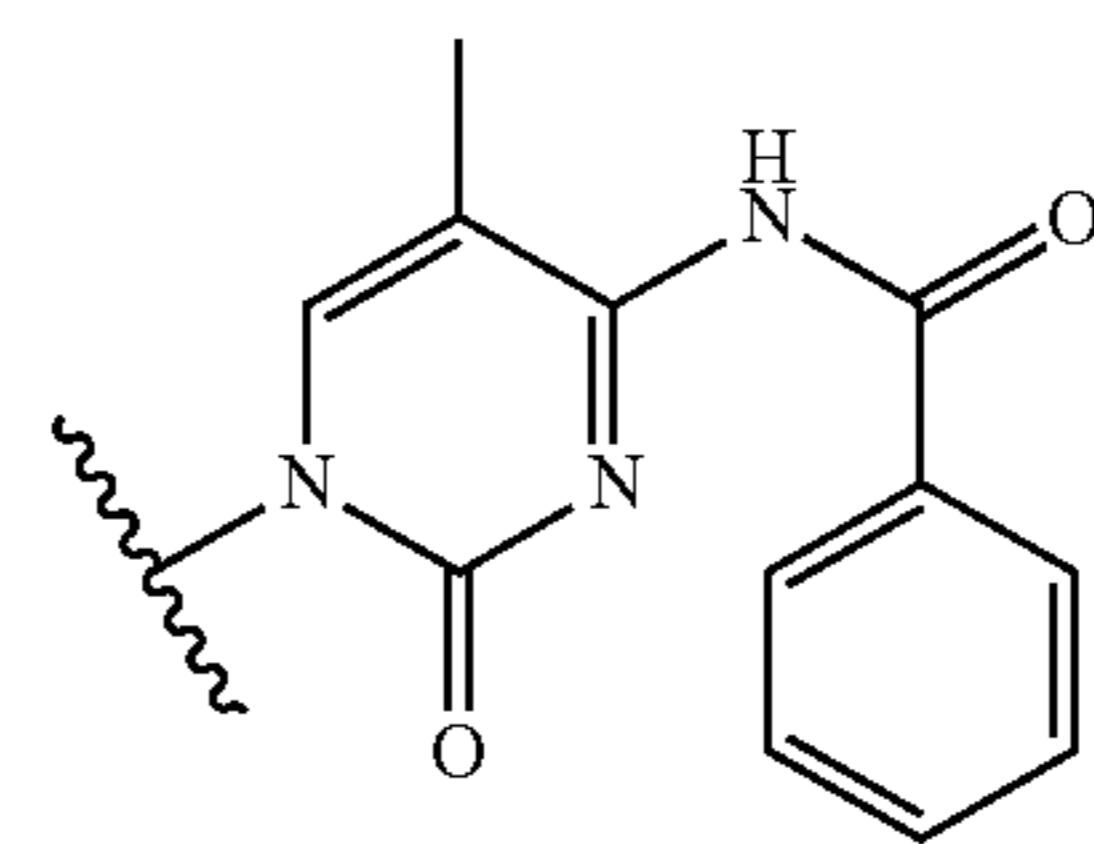
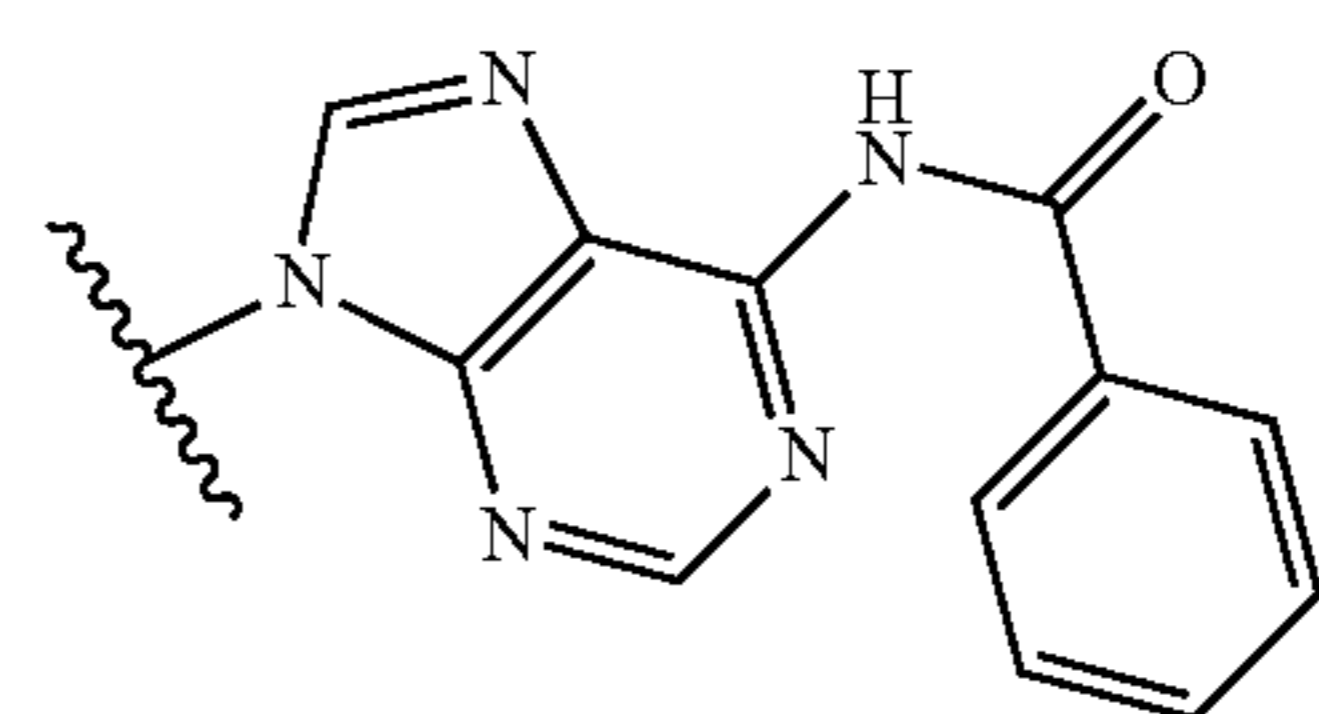
(A10)



wherein n is an integer from 10 to 40, R^1 is a support-medium, and R^4 is, independently for each occurrence, selected from the group consisting of:



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(PA)

(PC)

(P5mC)

(DPG)

(U)

(T)

(I)

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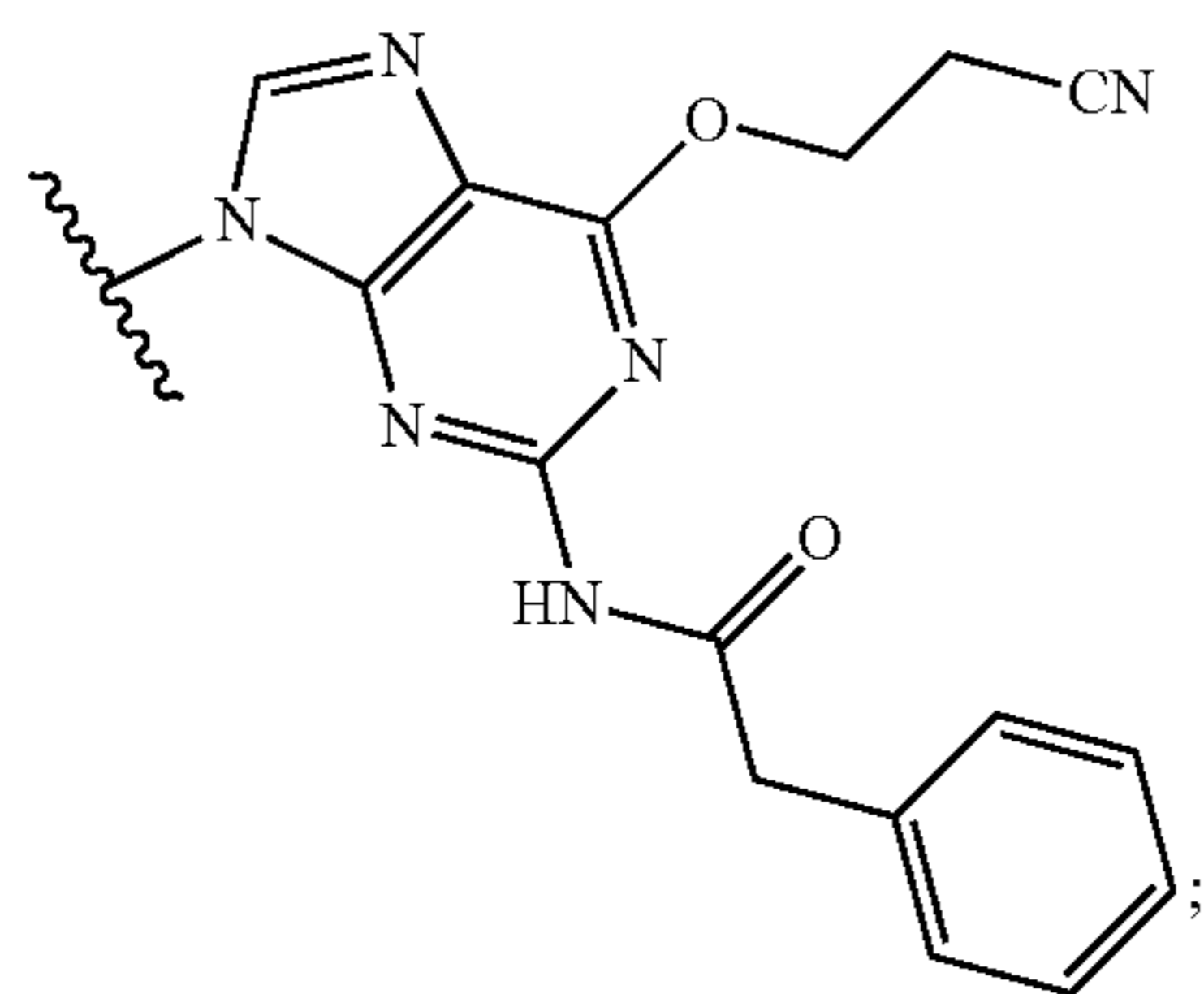
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37

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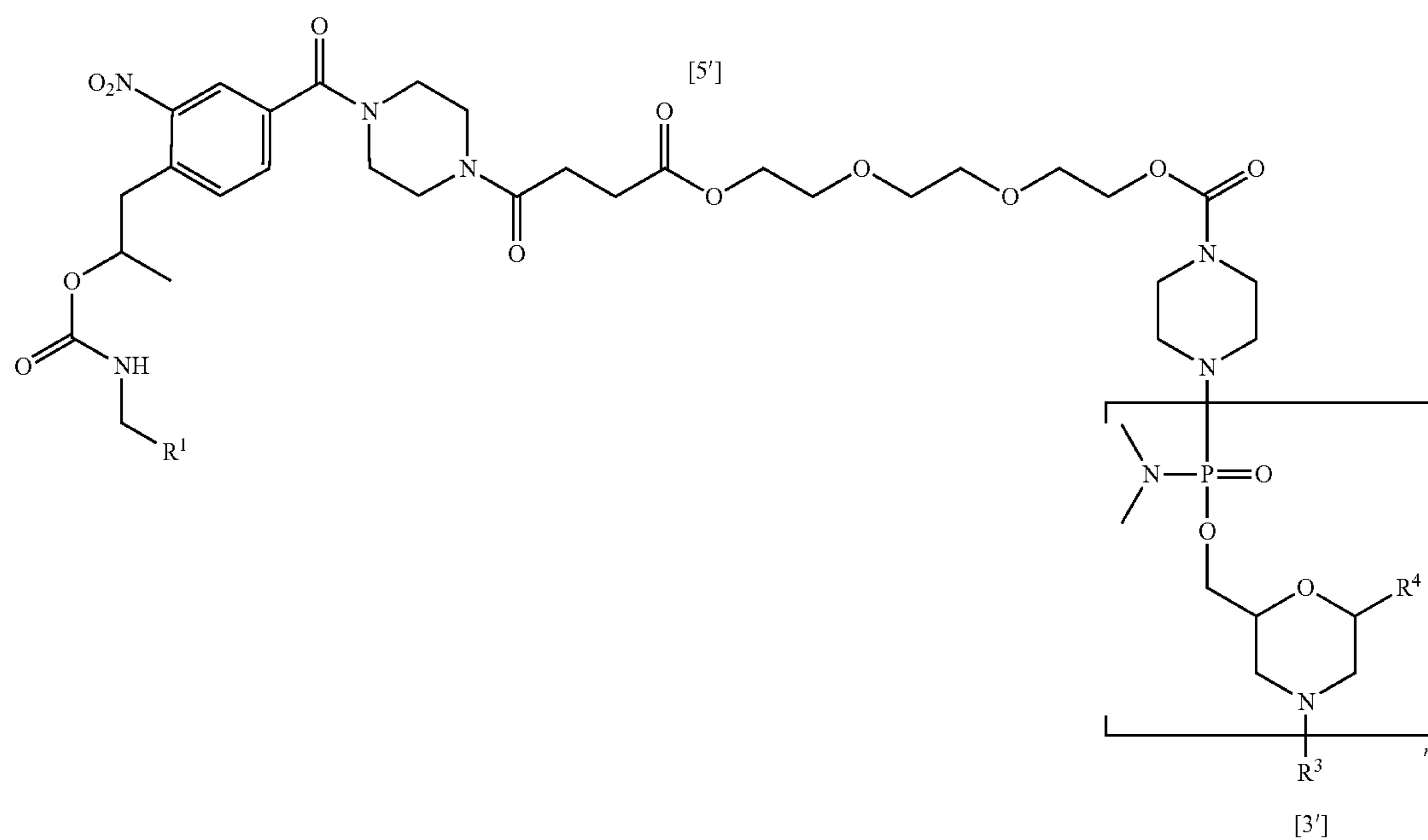


(PG)

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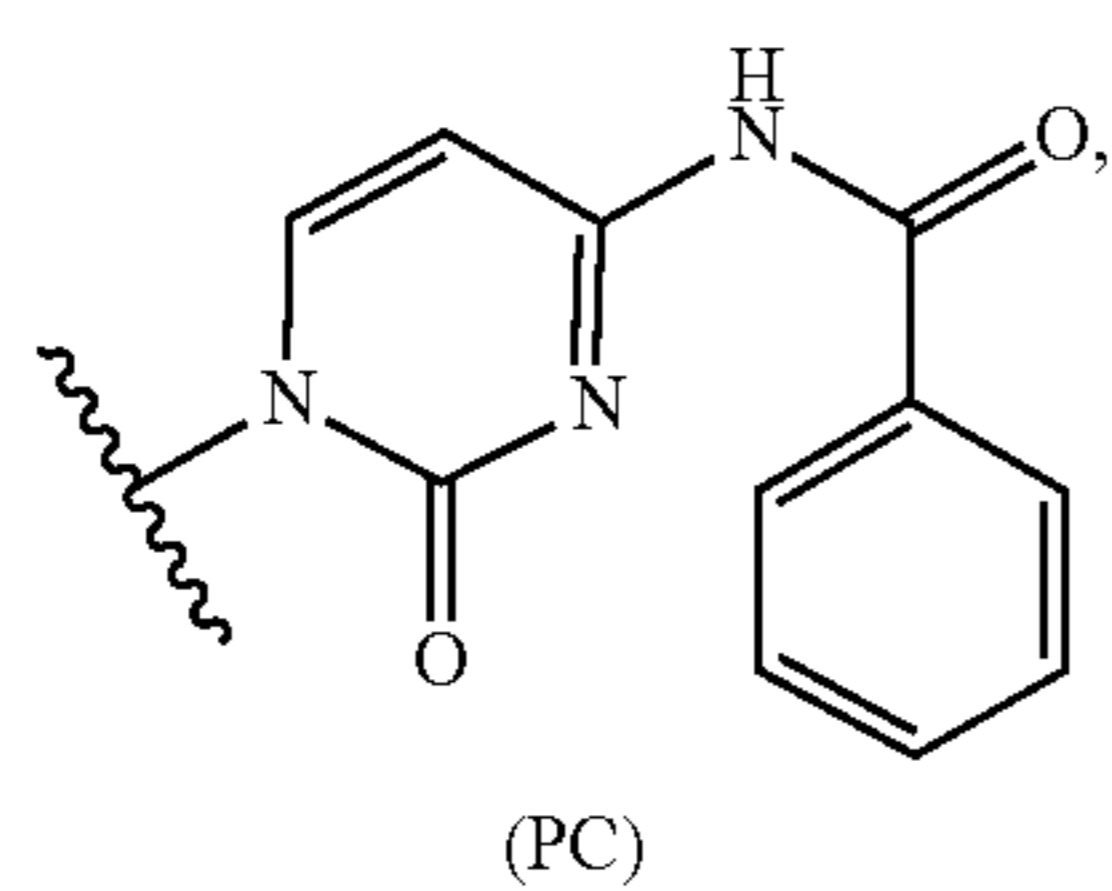
wherein the process comprises contacting a compound of ¹⁵ Formula (A9):



(A9)

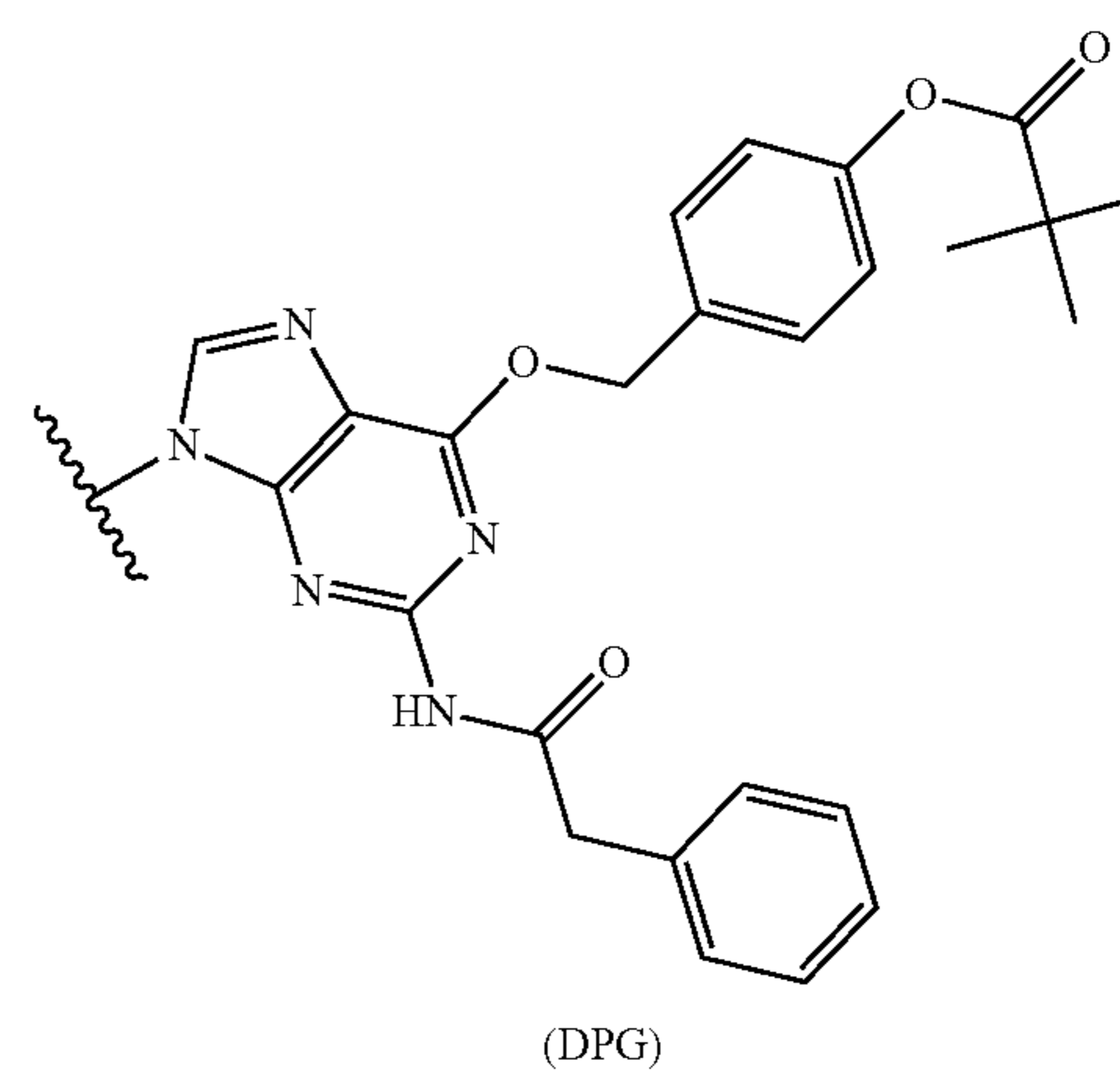
[3'] ;

wherein n is an integer from 10 to 40, R^1 is a support-⁴⁵ medium, R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R^4 is, independently for each occurrence, selected from the group consisting of:⁵⁰



(PC)

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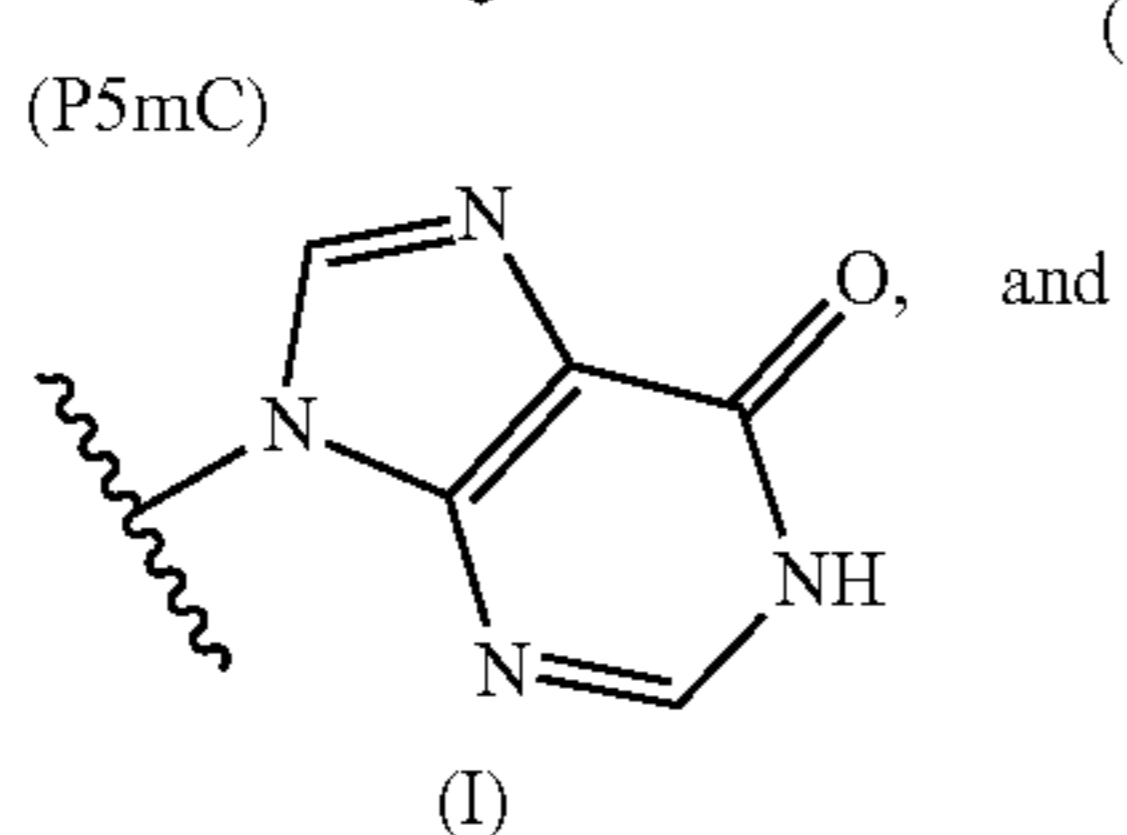
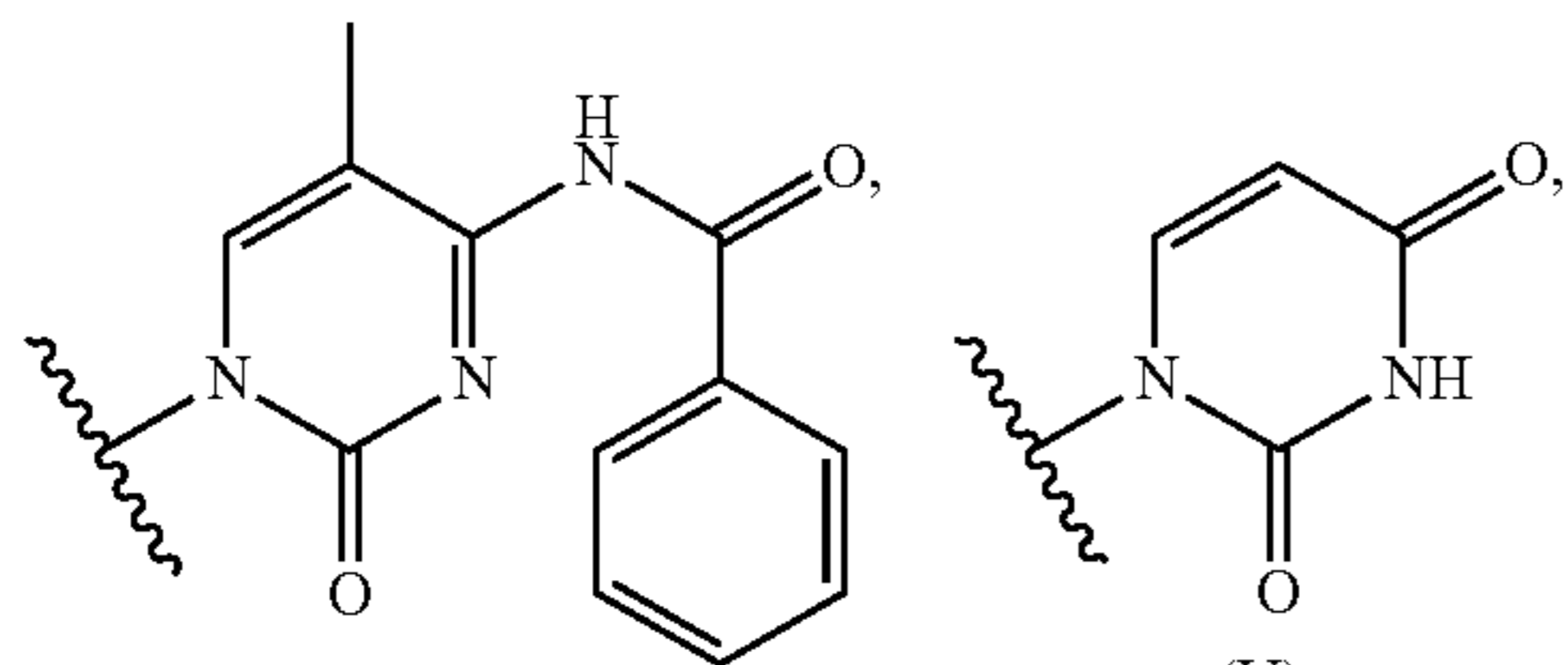
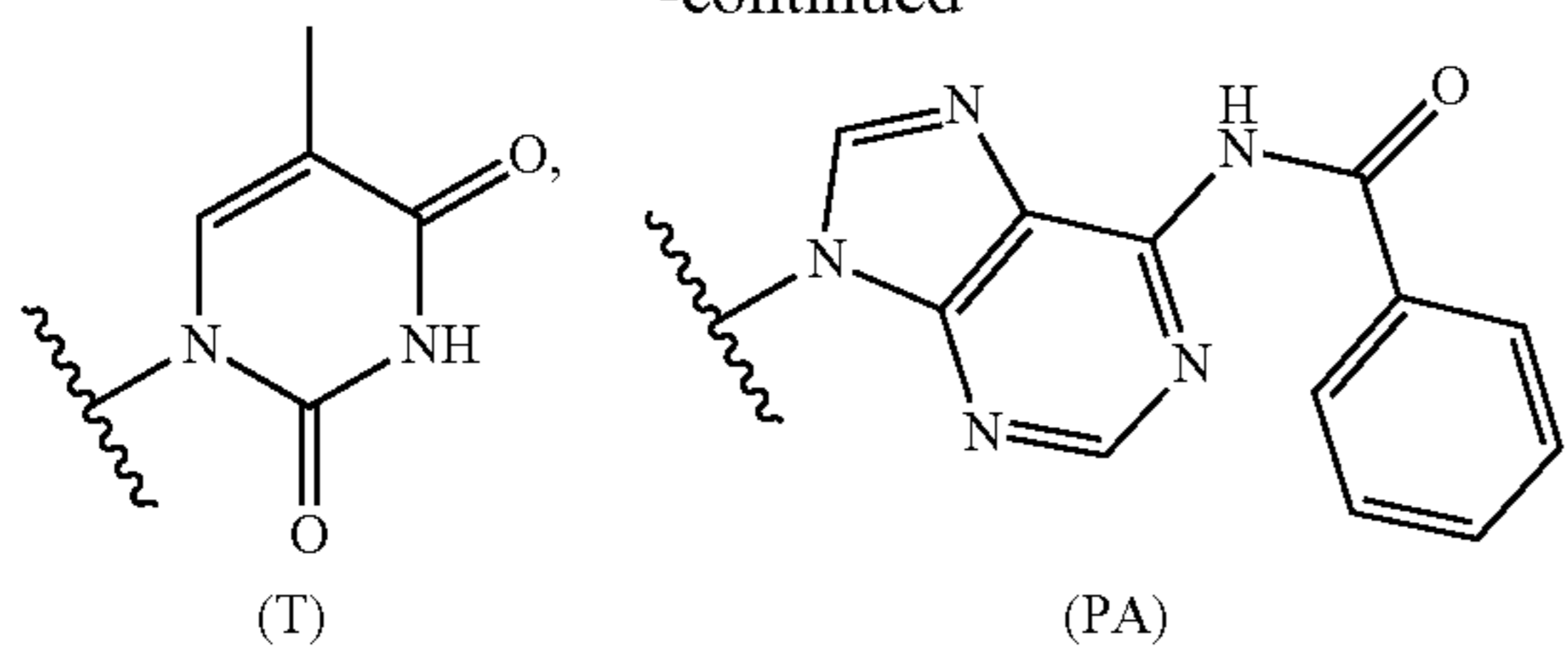


(DPG)

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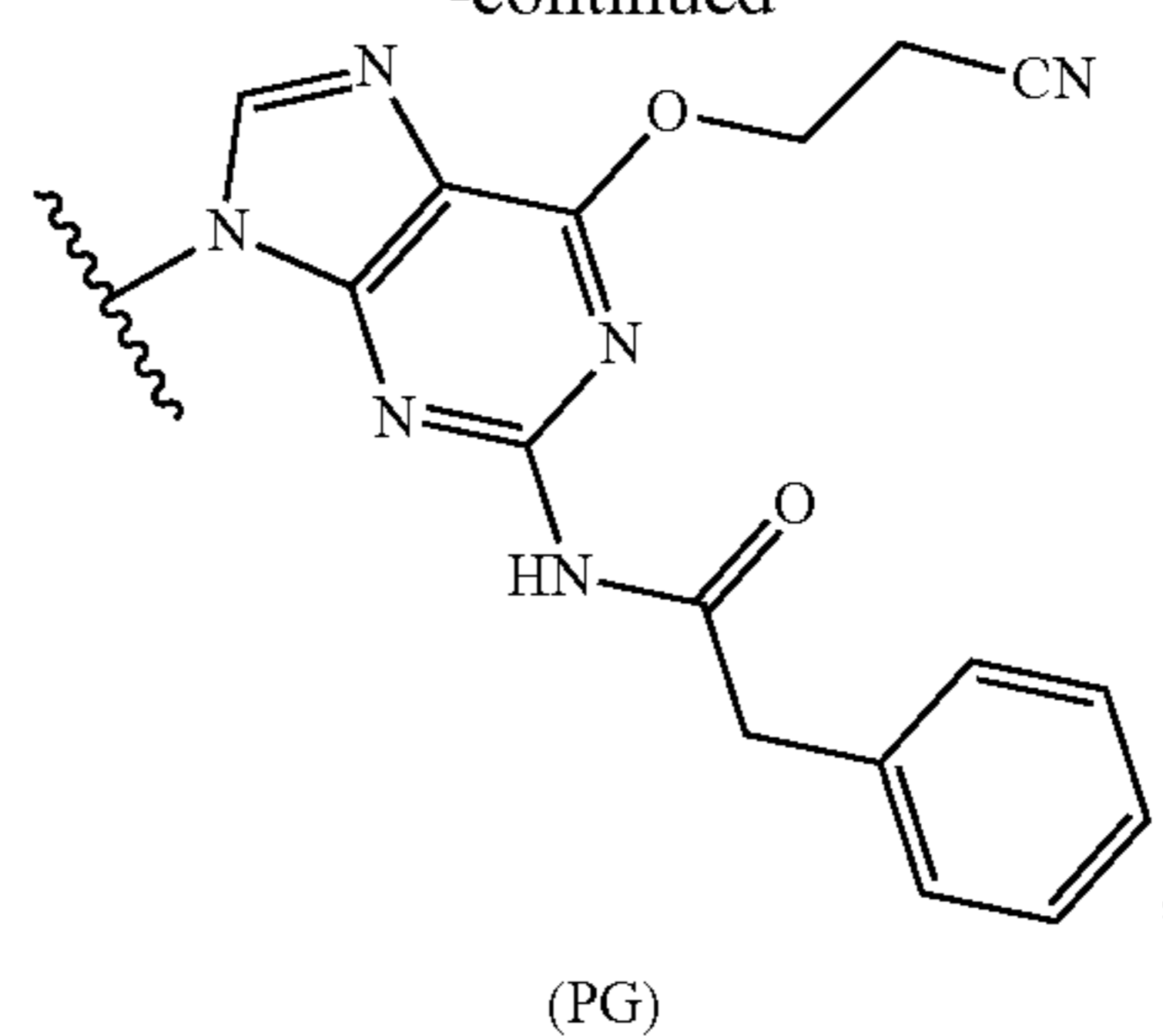
39

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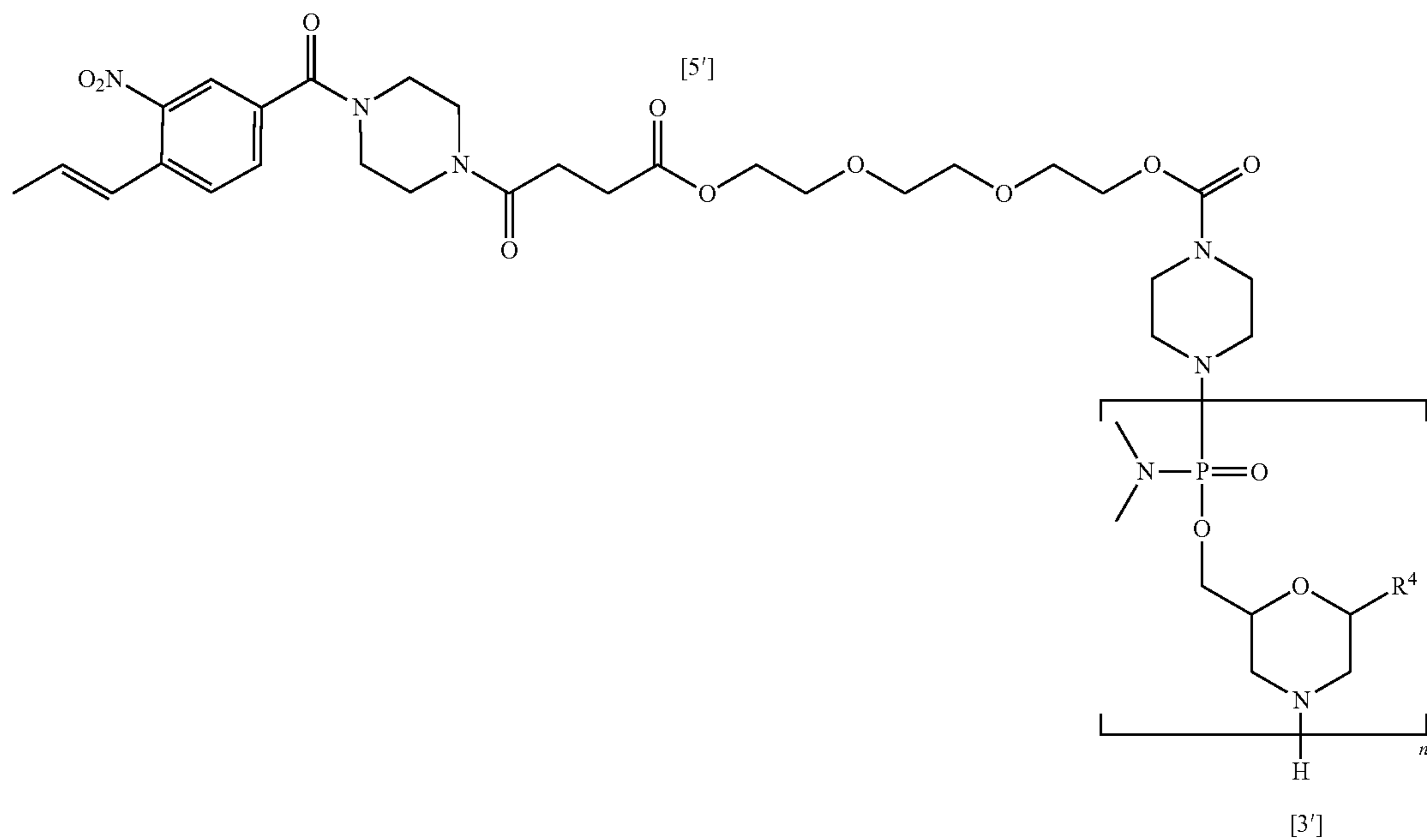
and

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with a deblocking agent to form a compound of Formula (A10).

In still another aspect, provided herein is a process for preparing a compound of Formula (A11):

(A11)

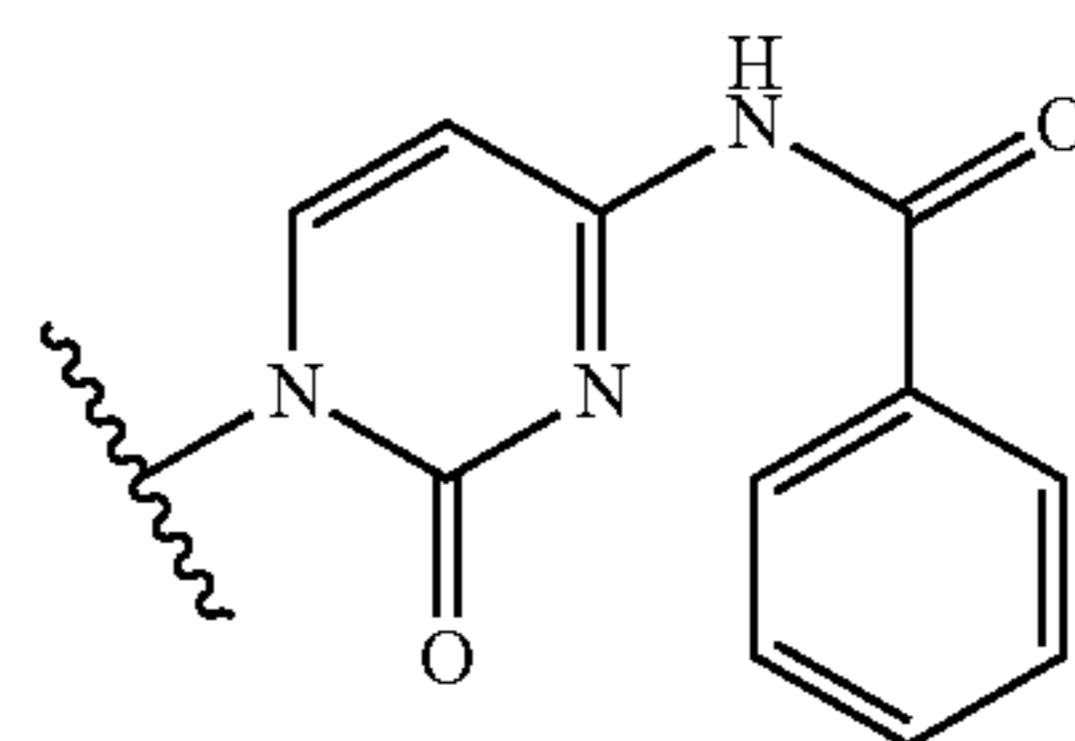


[3'] ;

wherein n is an integer from 10 to 40, and R⁴ is, for each occurrence independently selected from the group consisting of:

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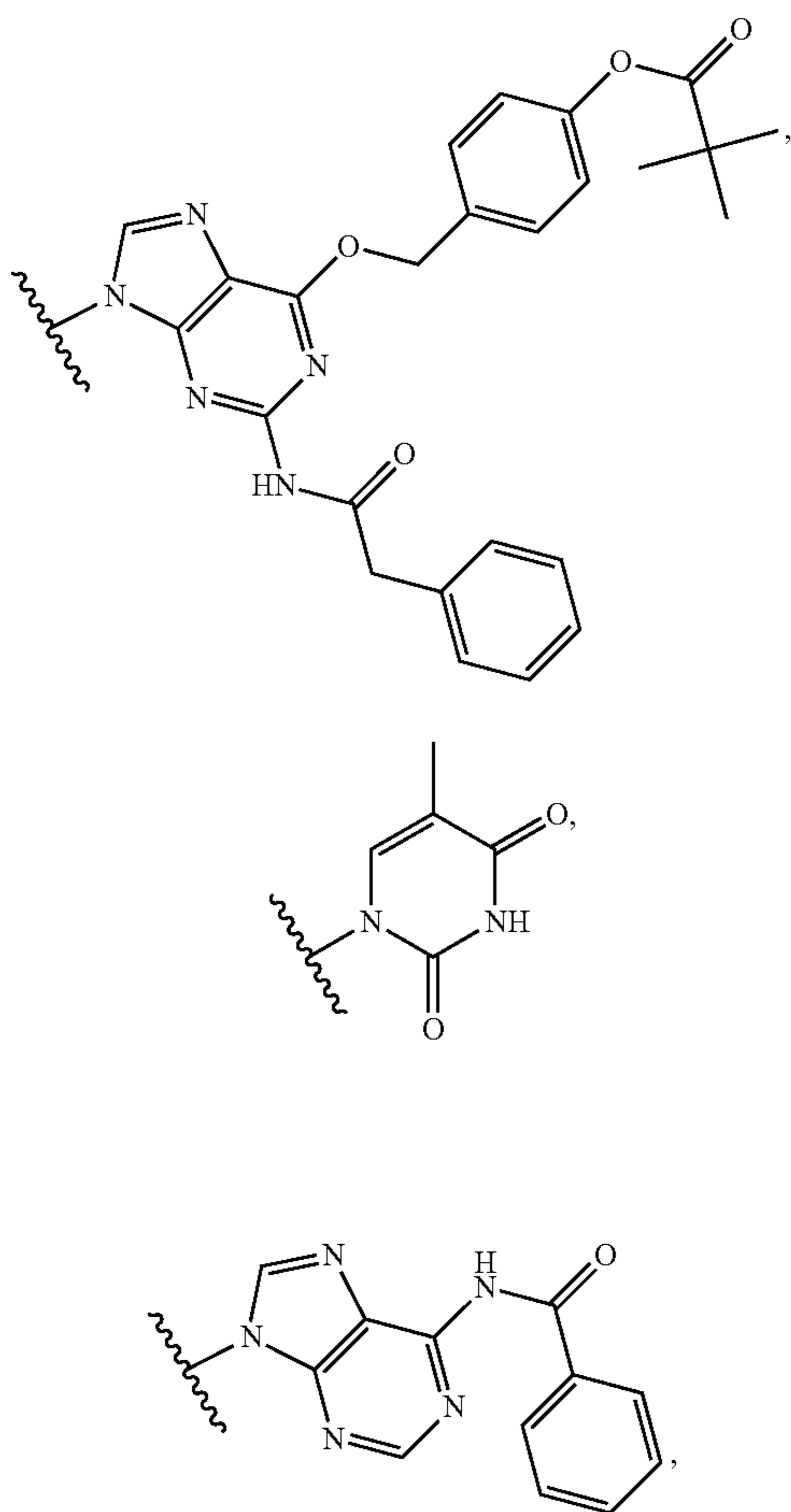
(PC)



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41

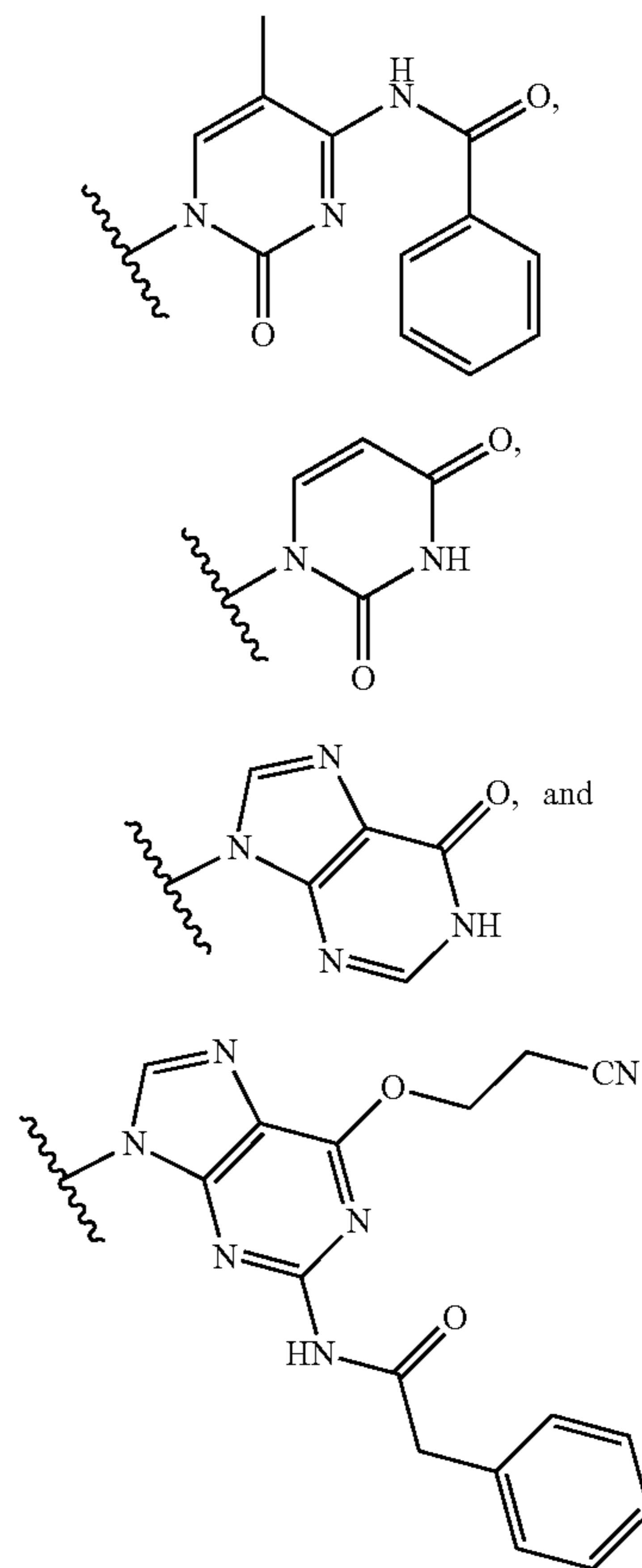
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42

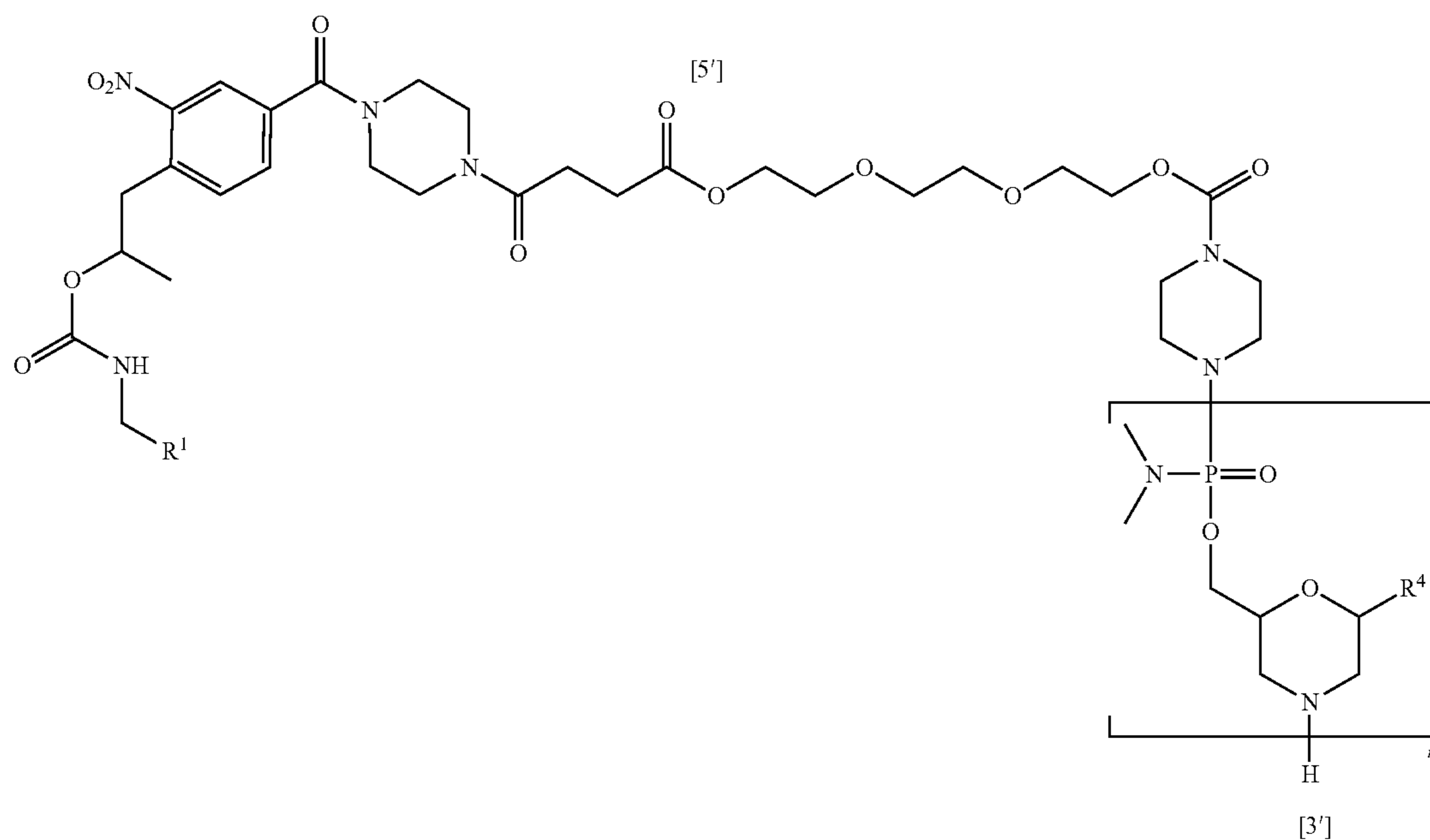
-continued

(P5mC)



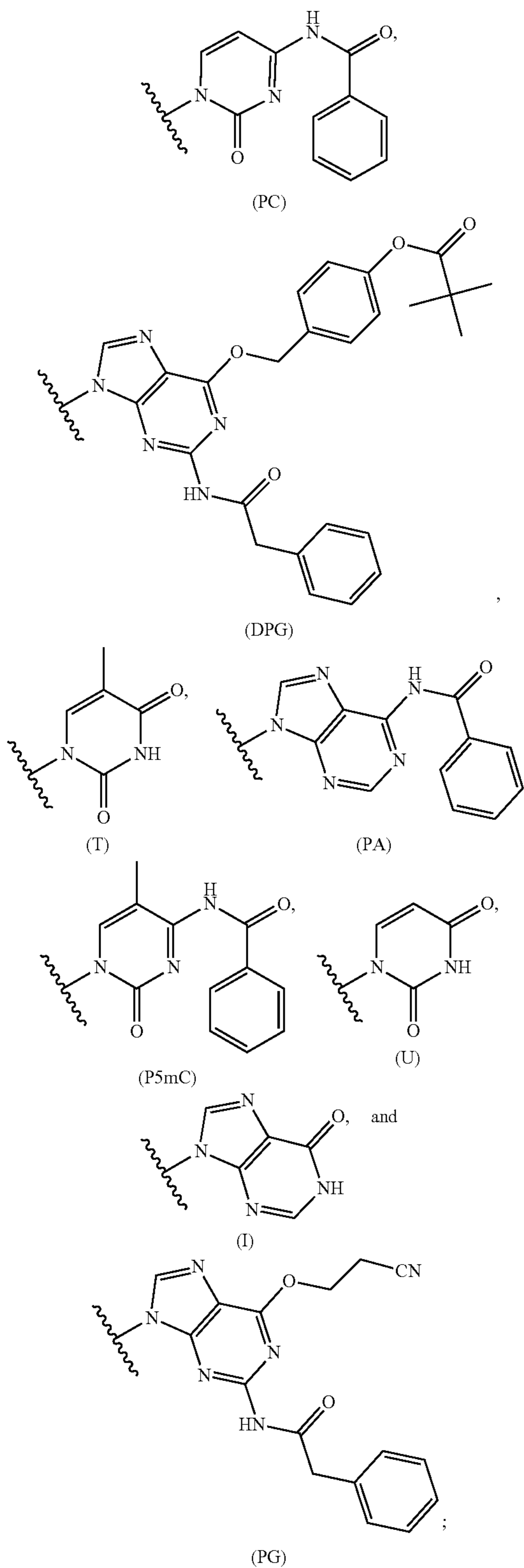
and
wherein the process comprises contacting the compound of
Formula (A10):

(A10)



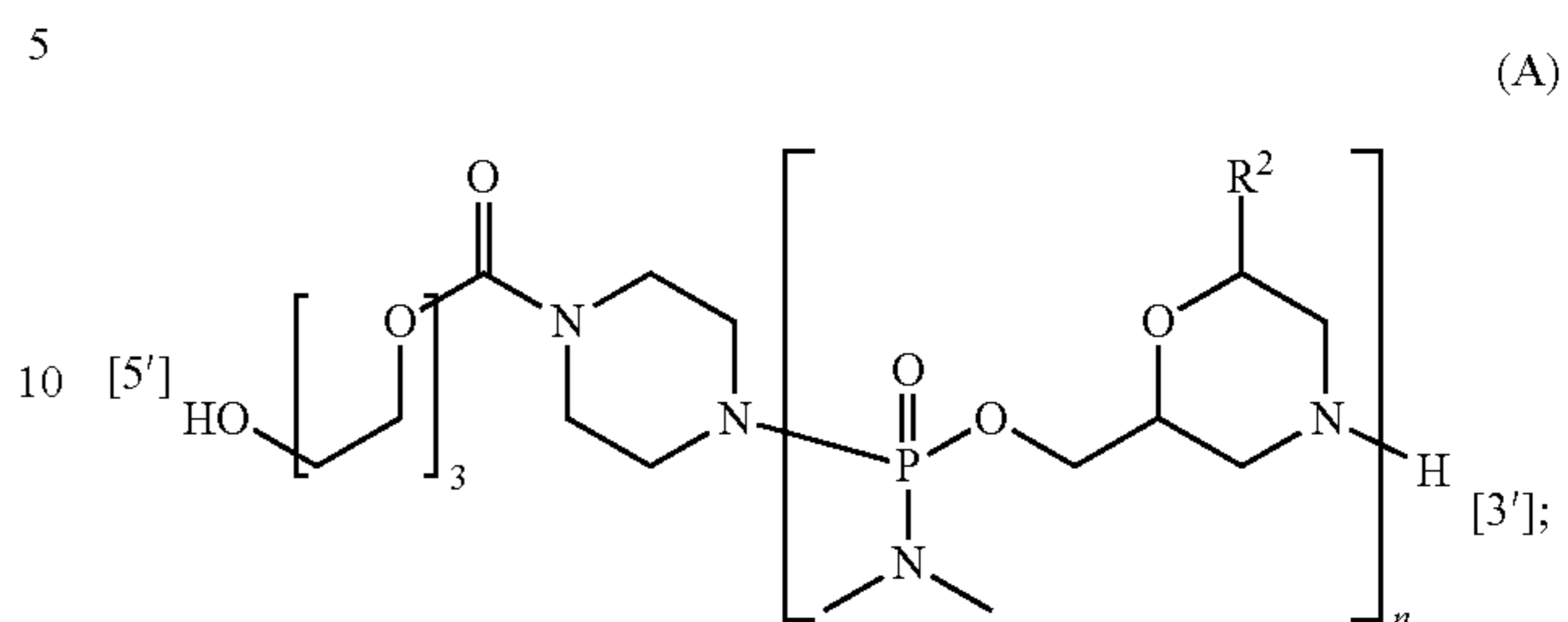
43

wherein n is an integer from 10 to 40, R¹ is a support-medium, and R⁴ is, independently for each occurrence, selected from the group consisting of:

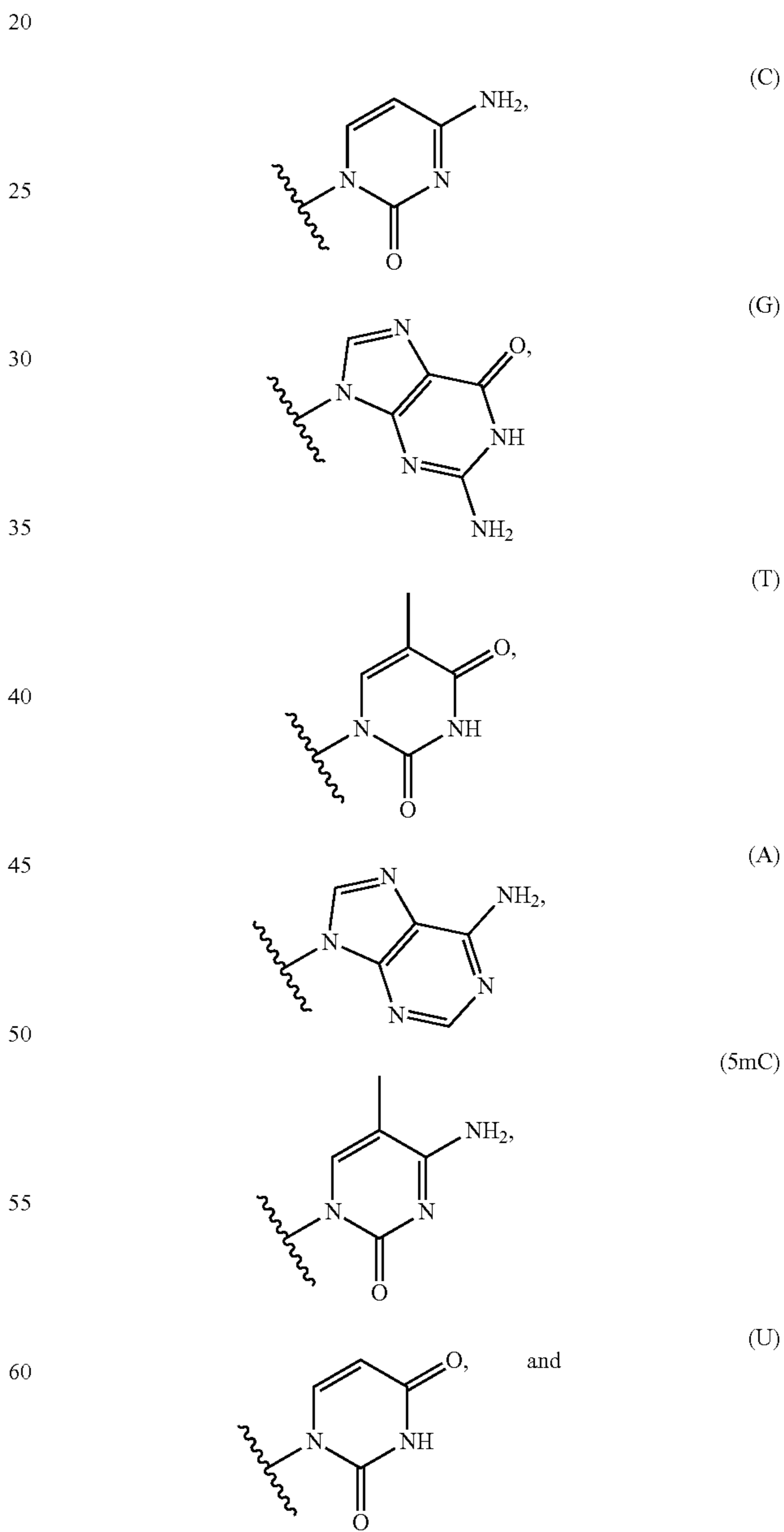


44

In another aspect, provided herein is a process for preparing an oligomeric compound of Formula (A):



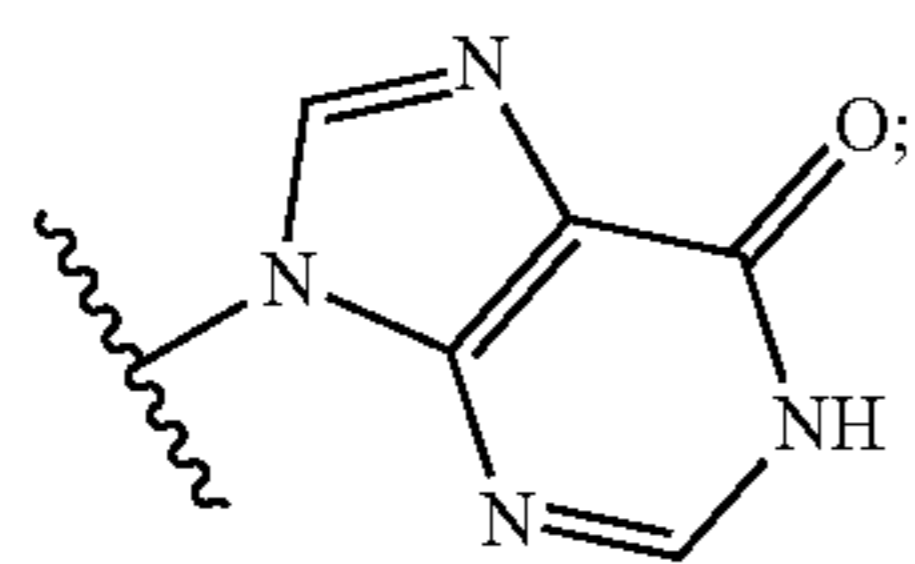
15 wherein n is an integer from 10 to 40, and each R² is, independently for each occurrence, selected from the group consisting of:



with a cleaving agent to form a compound of Formula (A11).

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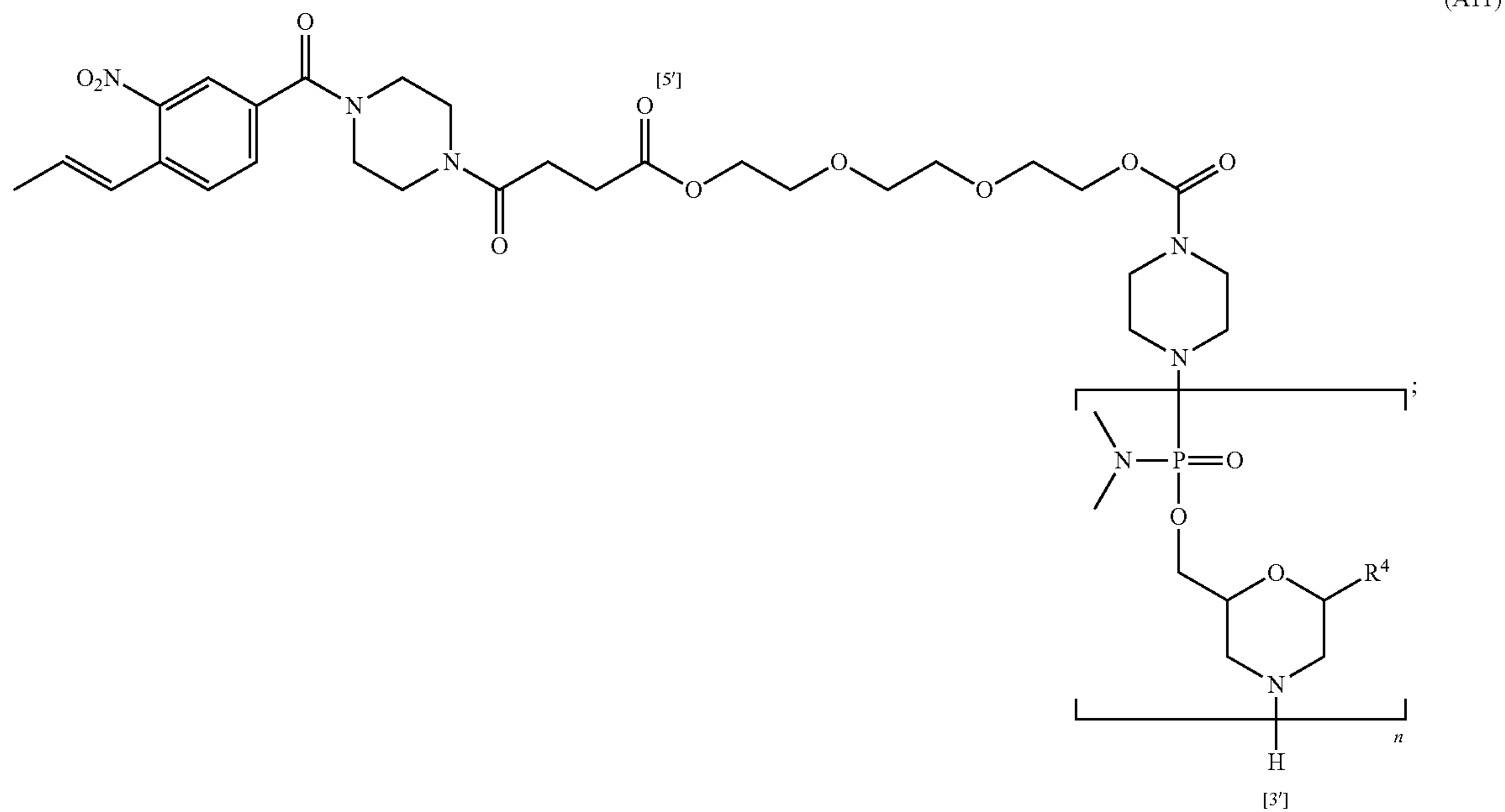


(I)

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46

wherein the process comprises contacting a compound of Formula (A11):

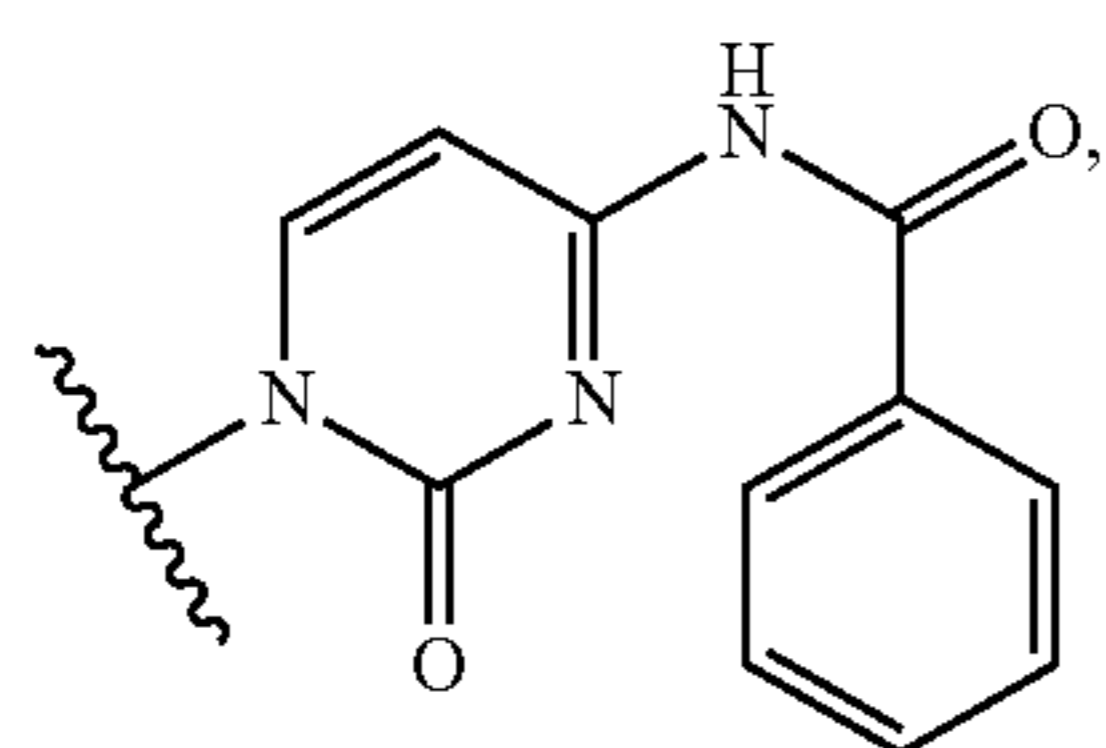


(A11)

wherein n is an integer from 10 to 40, and R⁴ is, independently for each occurrence, selected from the group consisting of:

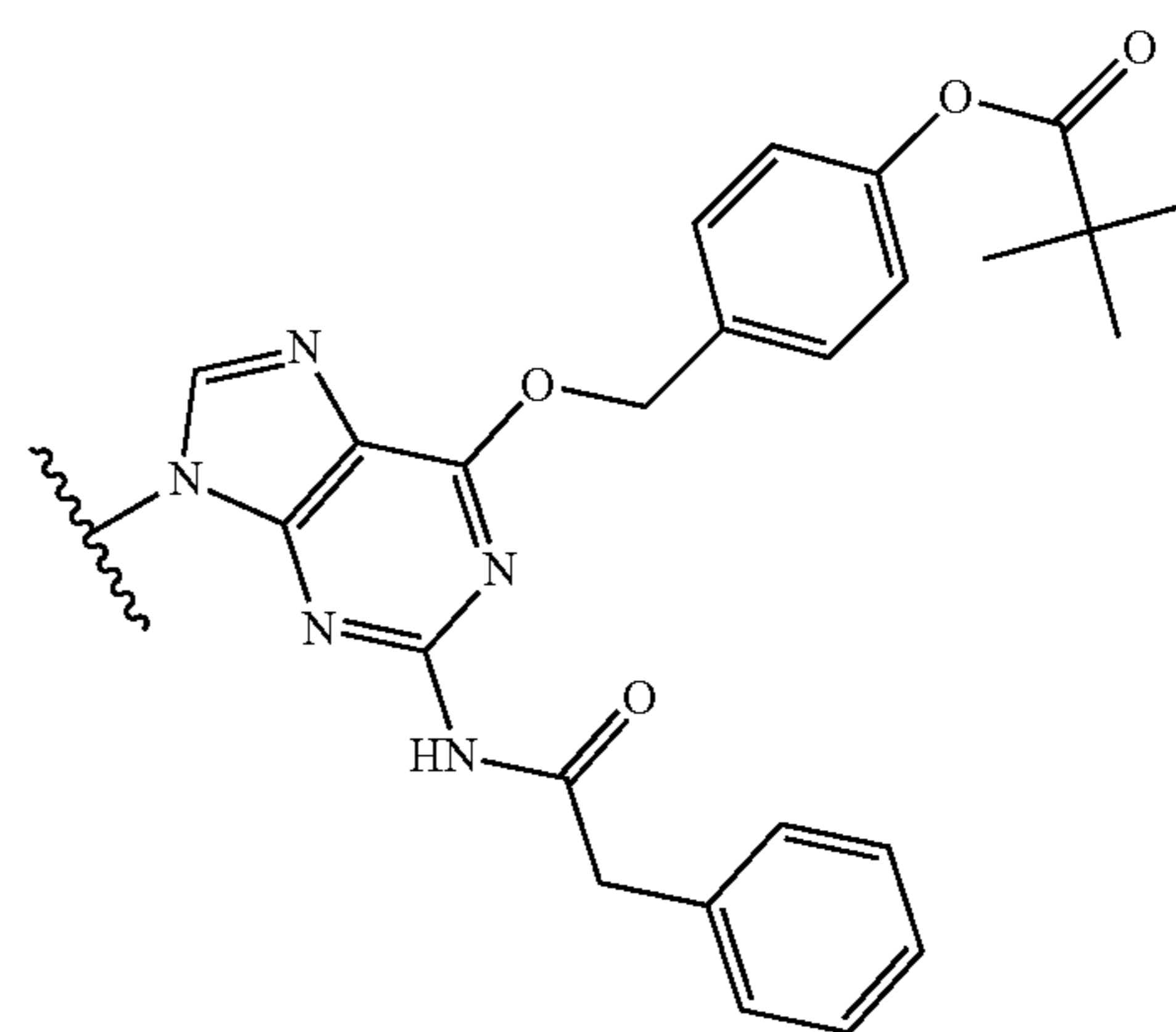
50

-continued



(PC)

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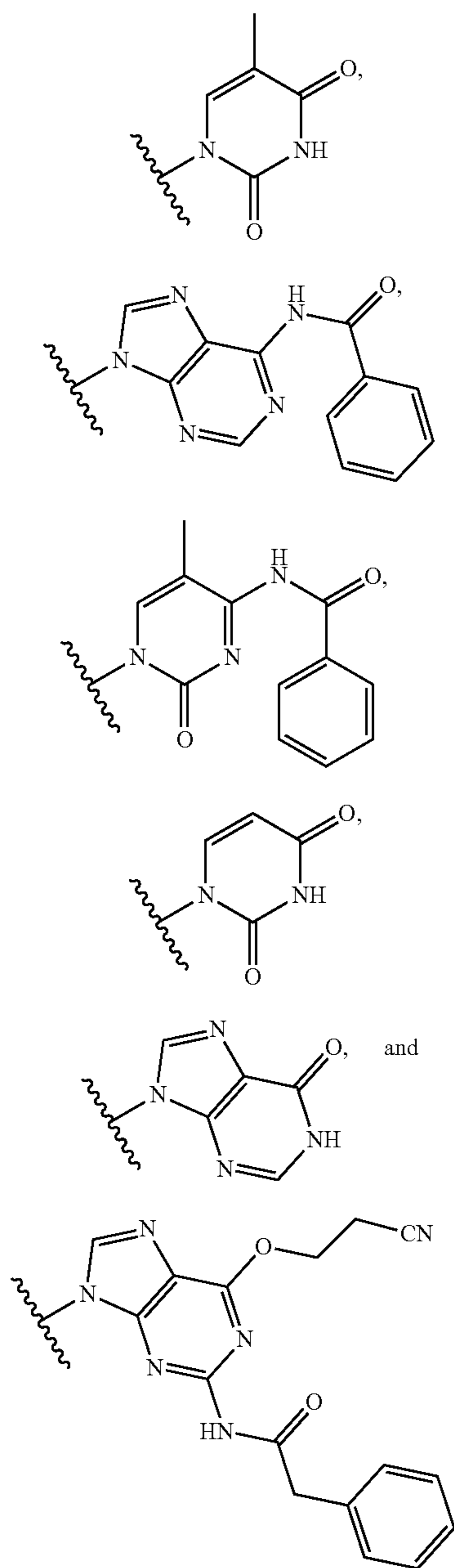
(DPG)

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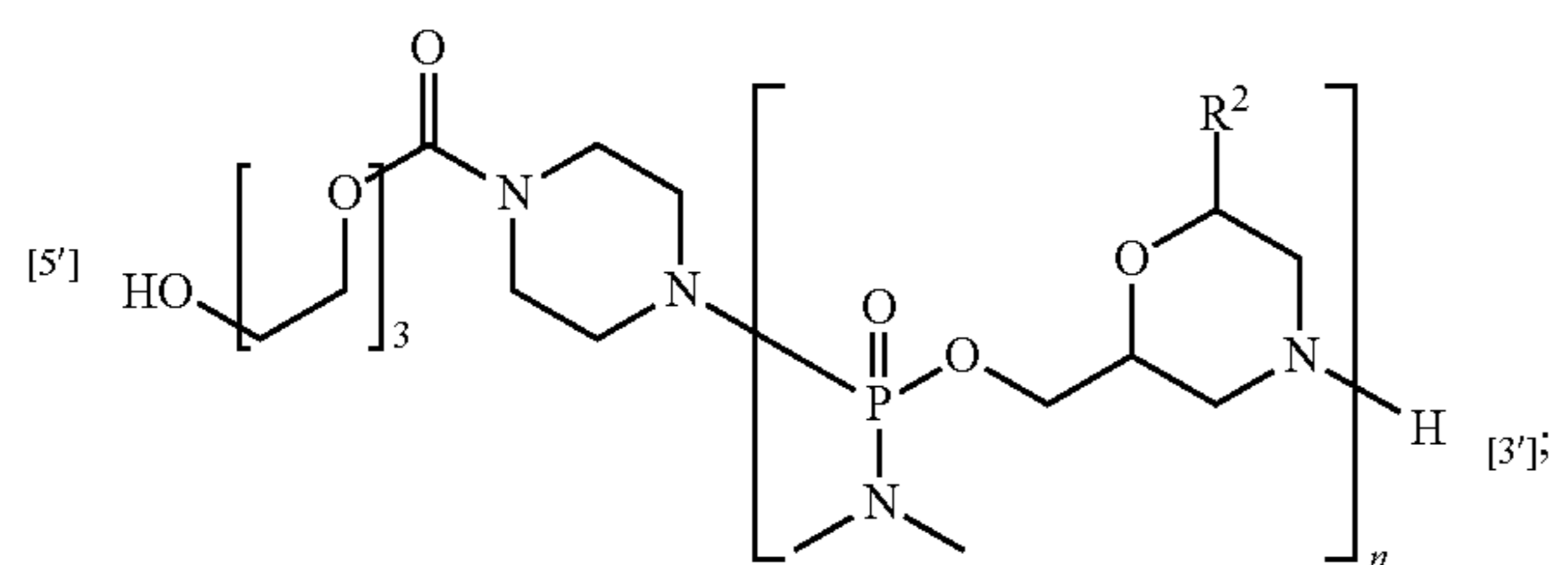
47

-continued



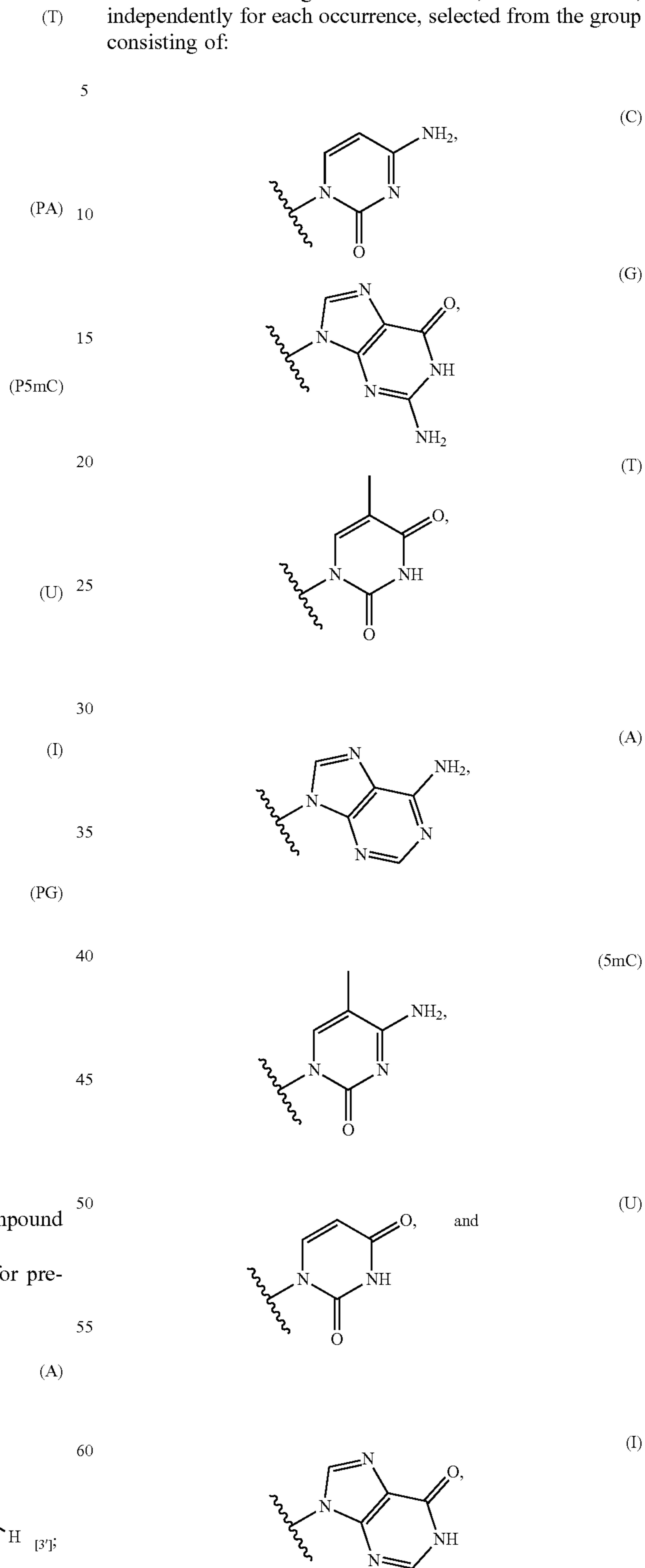
with a deprotecting agent to form the oligomeric compound of Formula (A).

In another aspect, provided herein is a process for preparing an oligomeric compound of Formula (A):



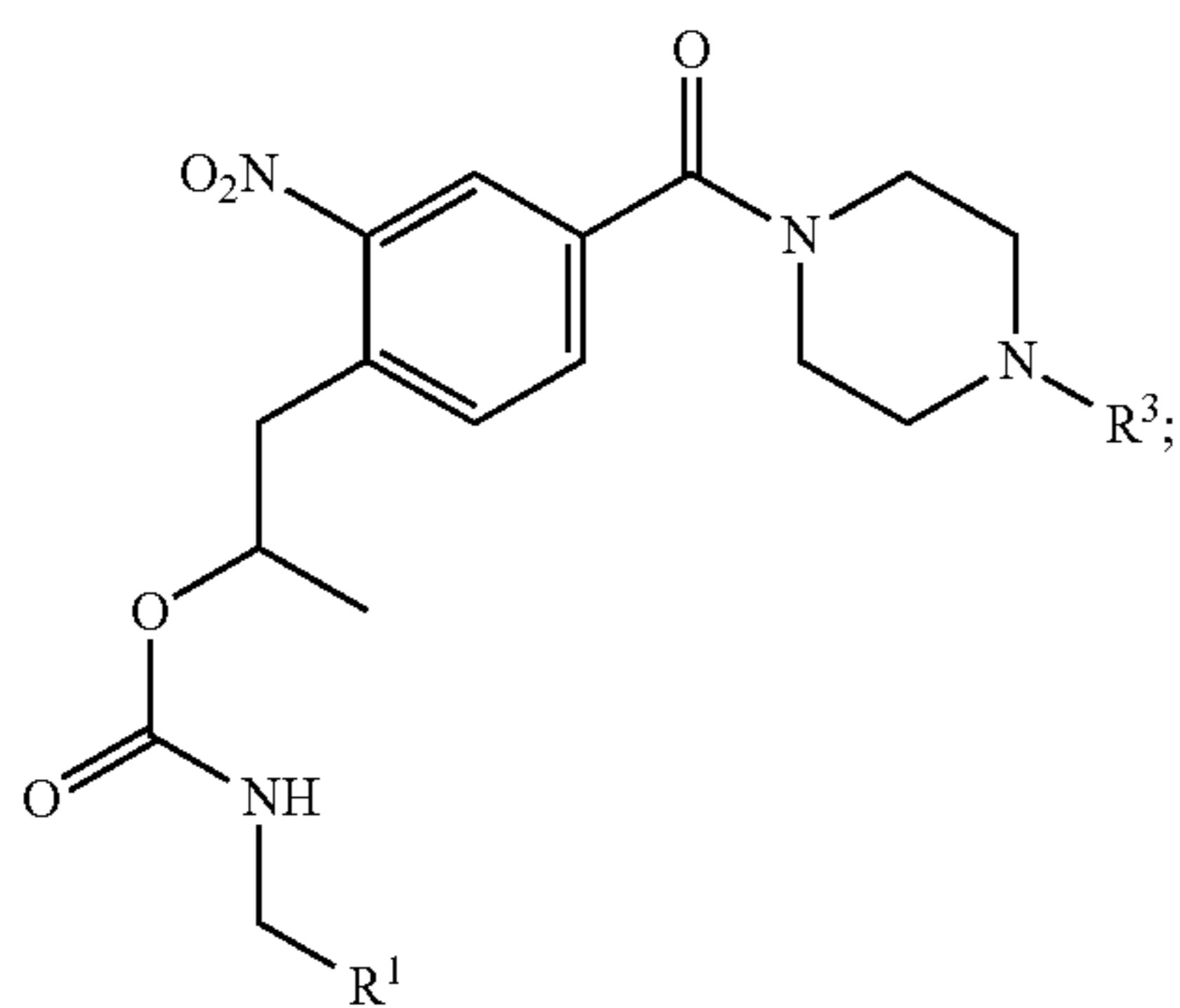
48

wherein n is an integer from 10 to 40, and each R² is, independently for each occurrence, selected from the group consisting of:



49

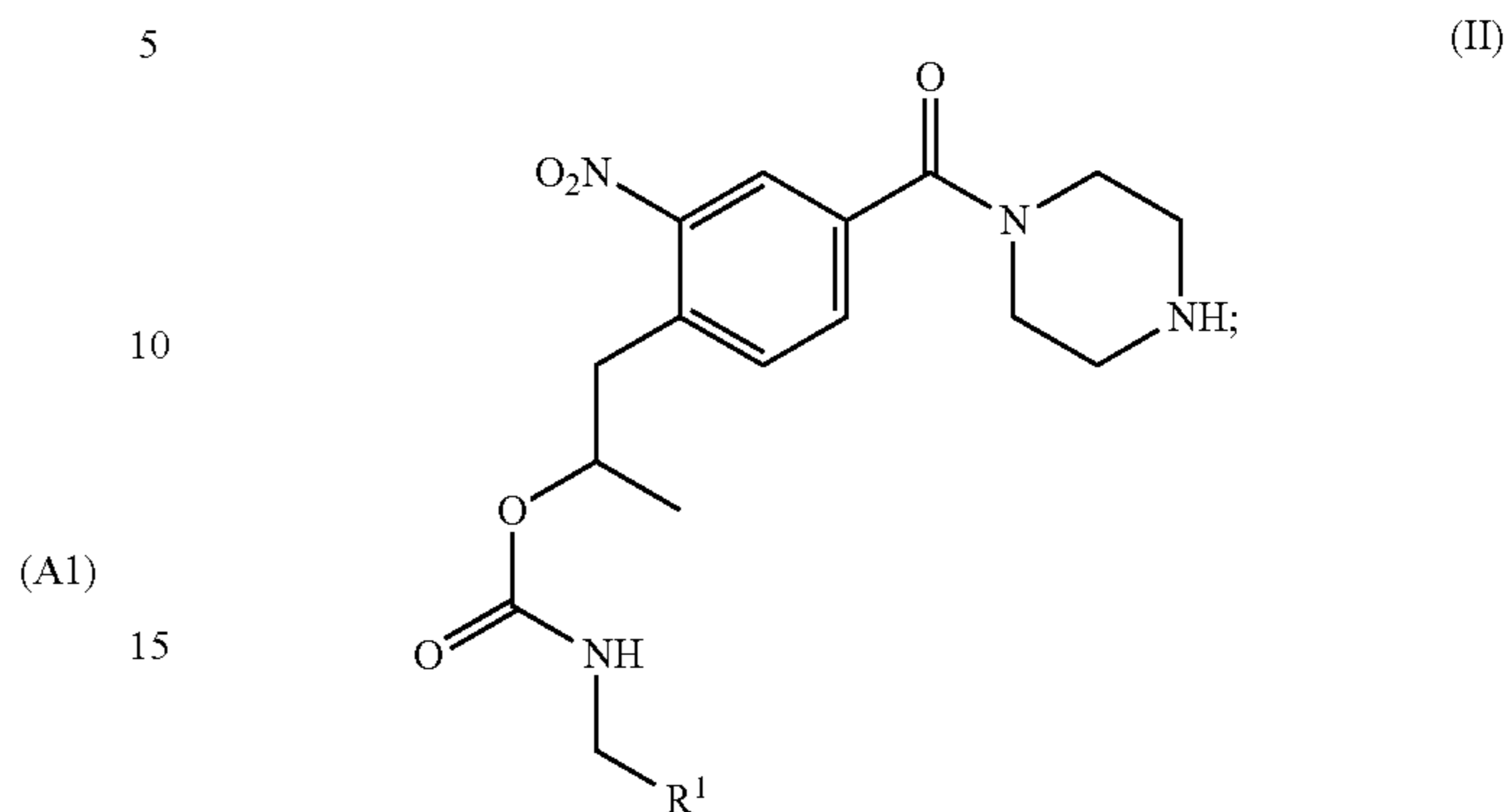
wherein the process comprises the sequential steps of:
 (a) contacting a compound of Formula (A1):



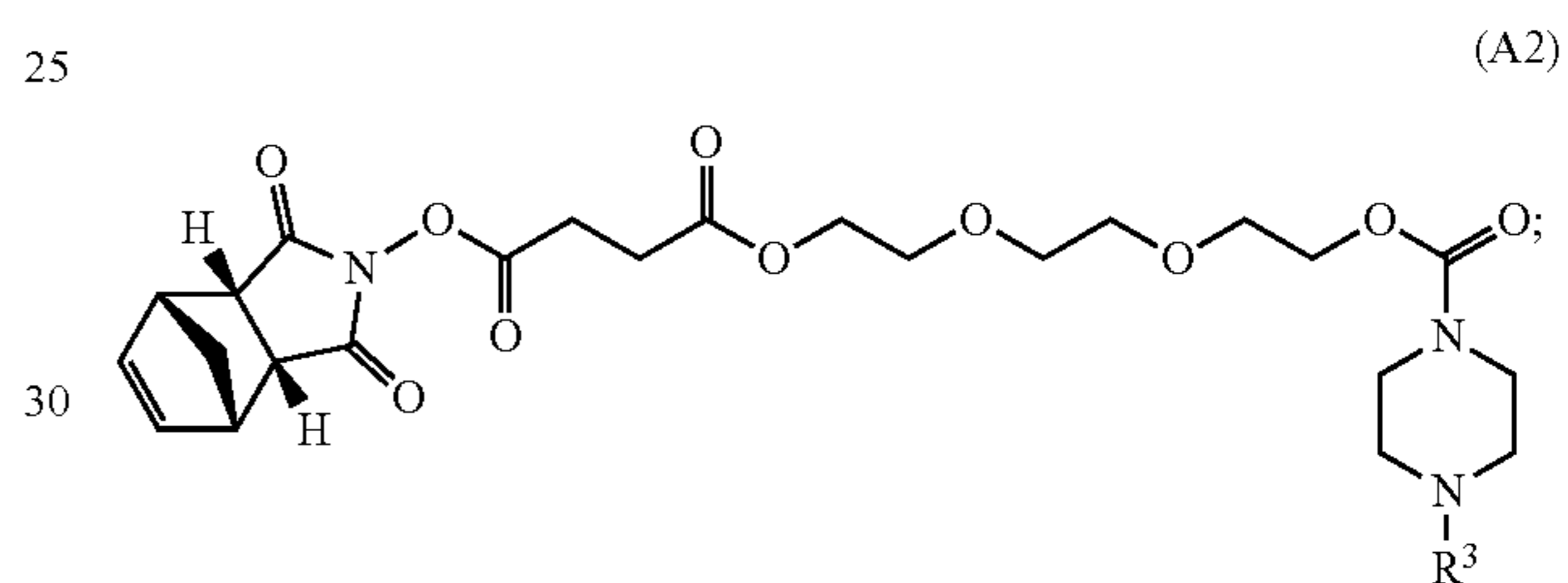
wherein R¹ is a support-medium and R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

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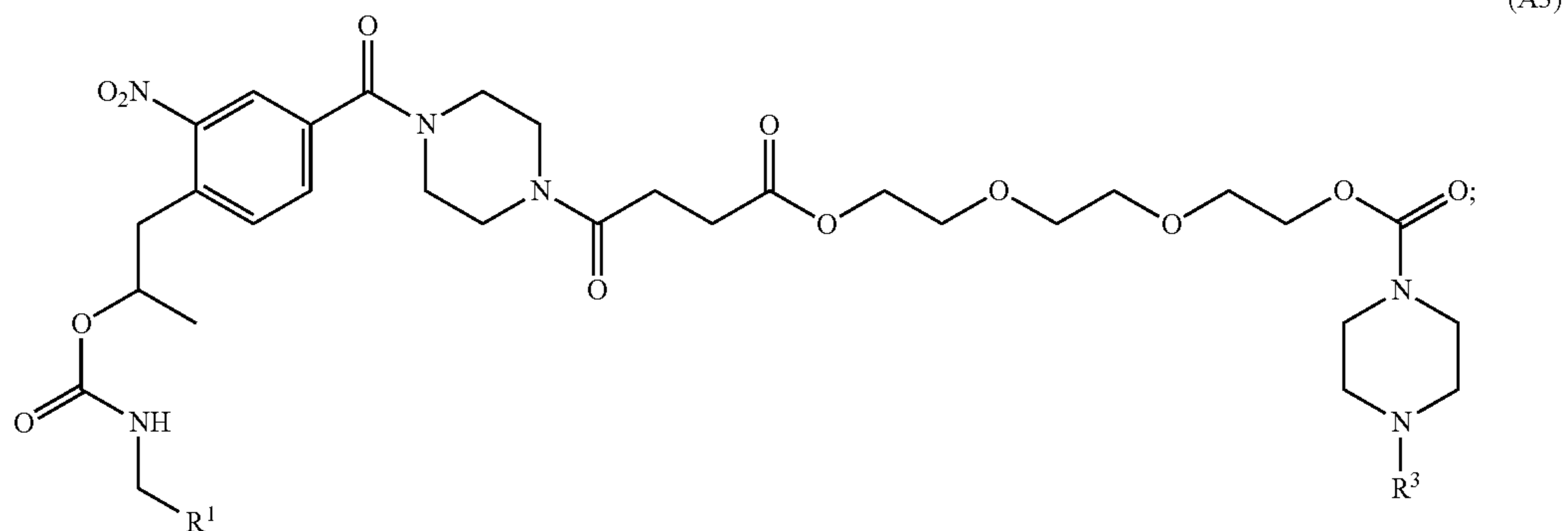
with a deblocking agent to form the compound of Formula (II):



wherein R¹ is a support-medium;
 (b) contacting the compound of Formula (II) with a compound of Formula (A2):



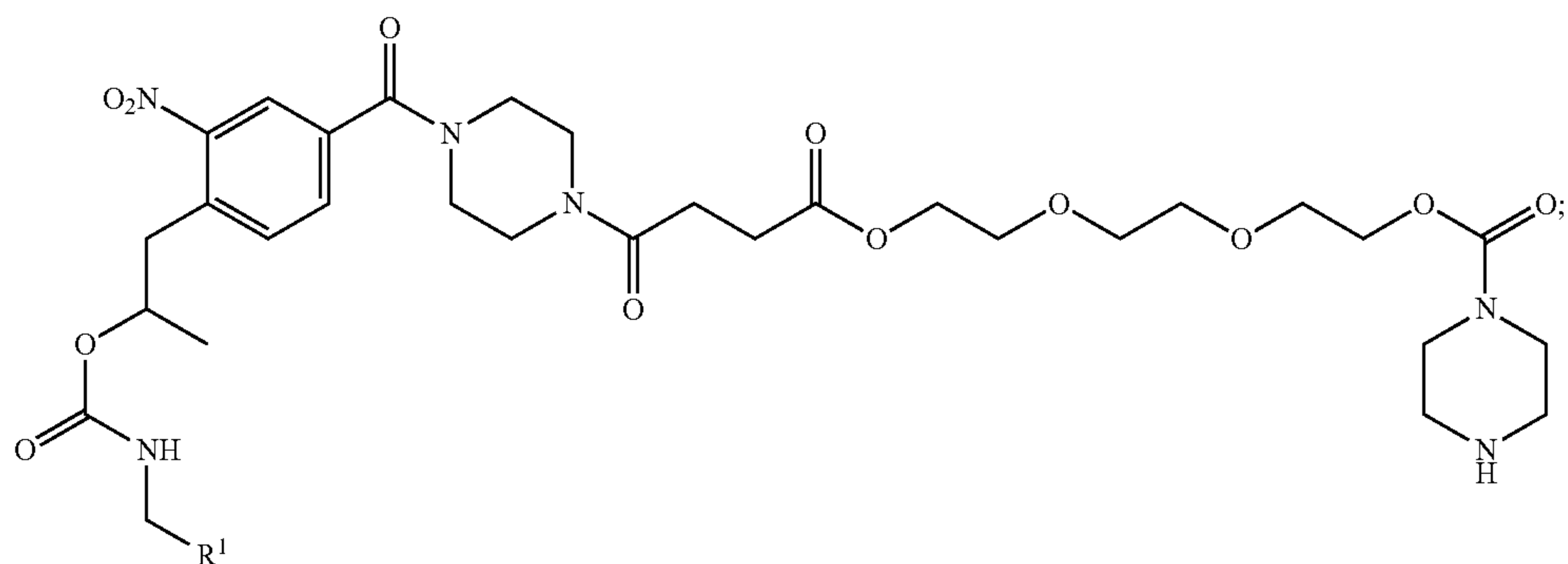
wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
 to form a compound of Formula (A3):



wherein R¹ is a support-medium, and R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
 (c) contacting the compound of Formula (A3) with a deblocking agent to form a compound of Formula (IV):

51

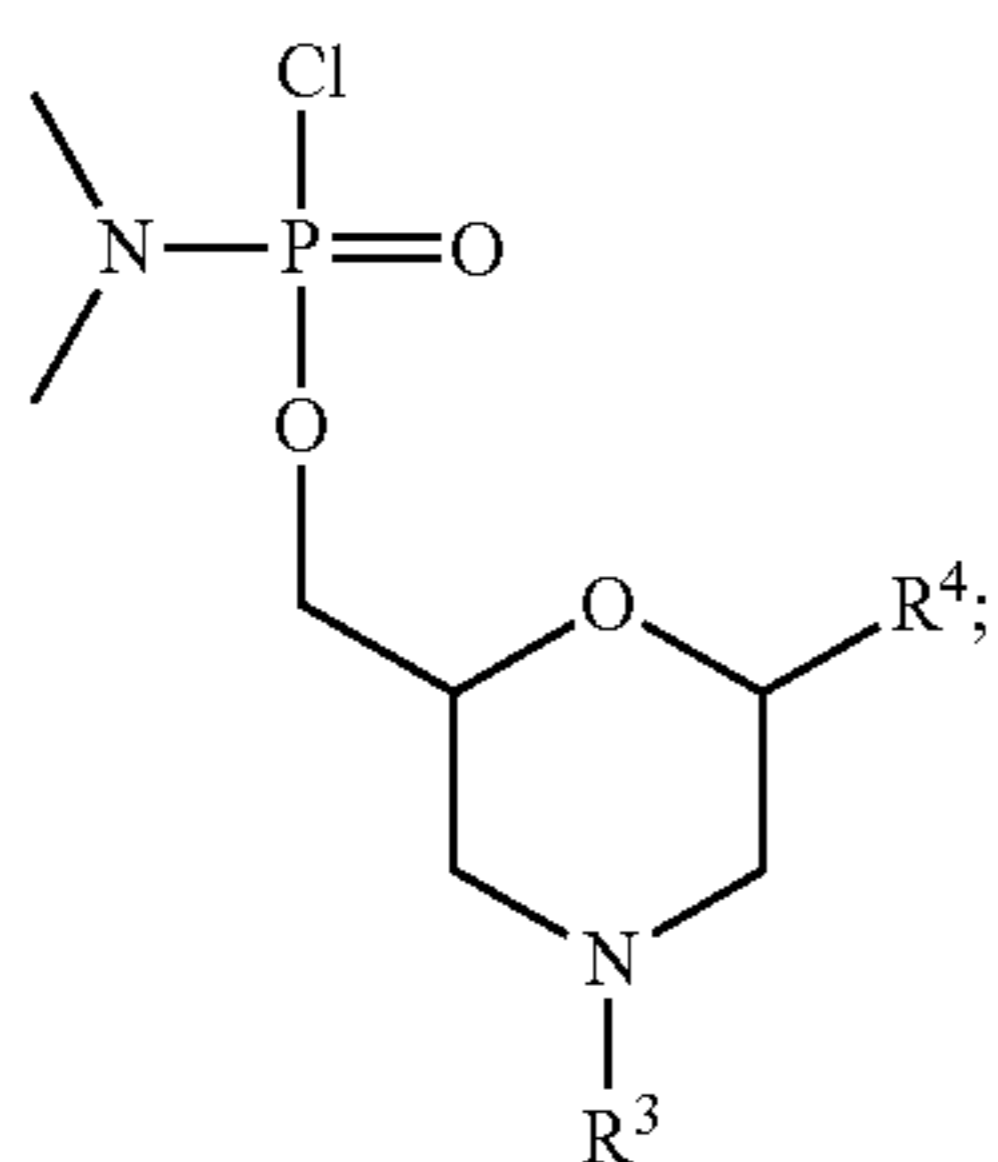
52



(IV)

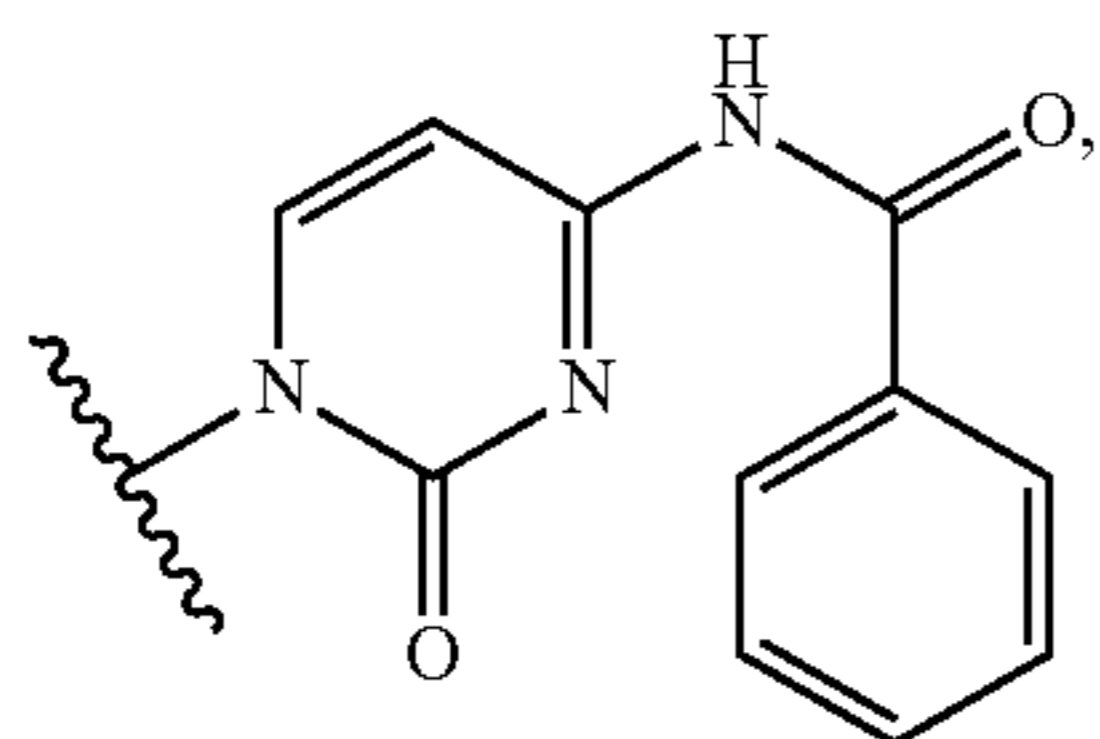
wherein R¹ is a support-medium;

(d) contacting the compound of Formula (IV) with a compound of Formula (A4):

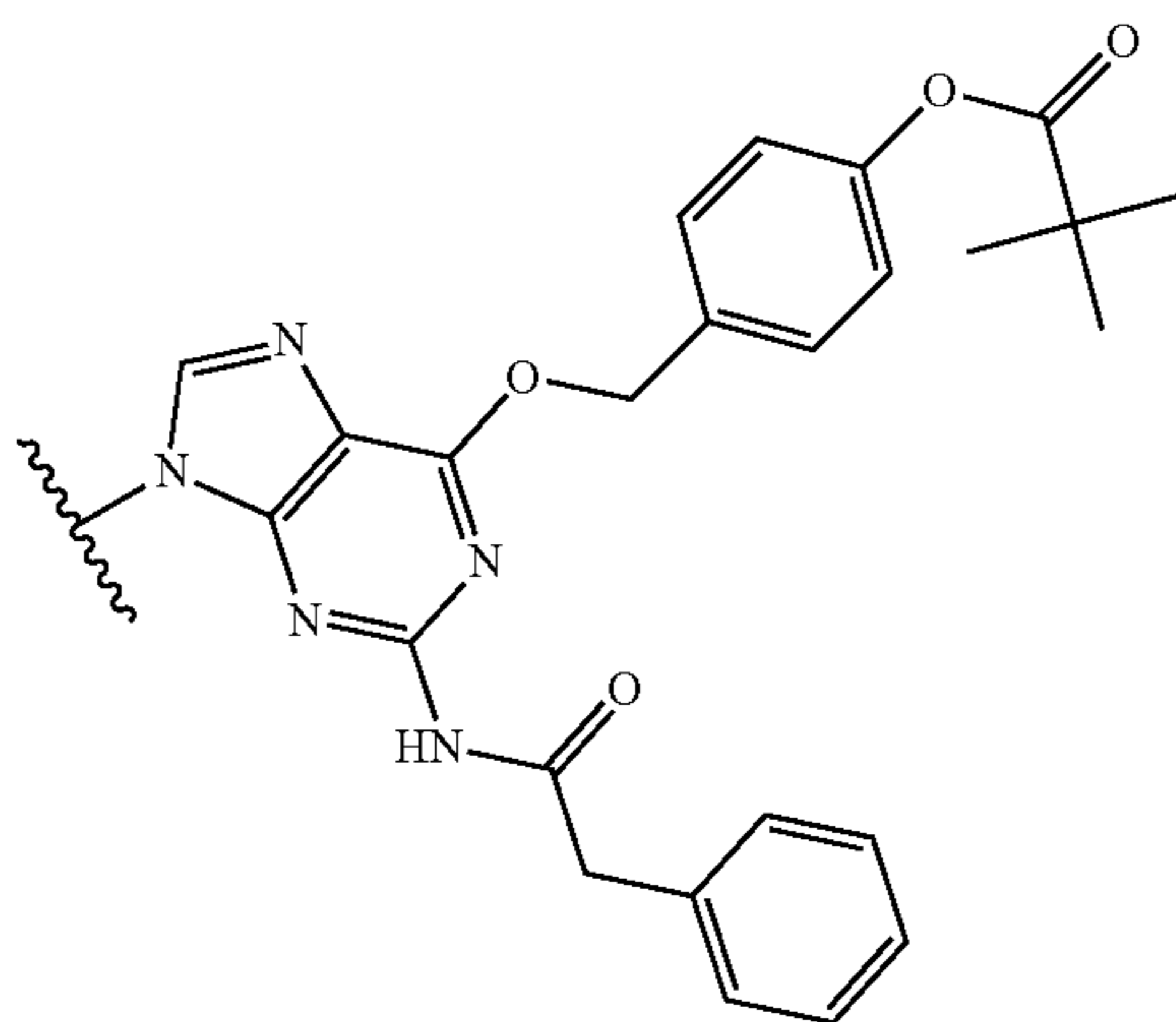


(A4) 25

wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

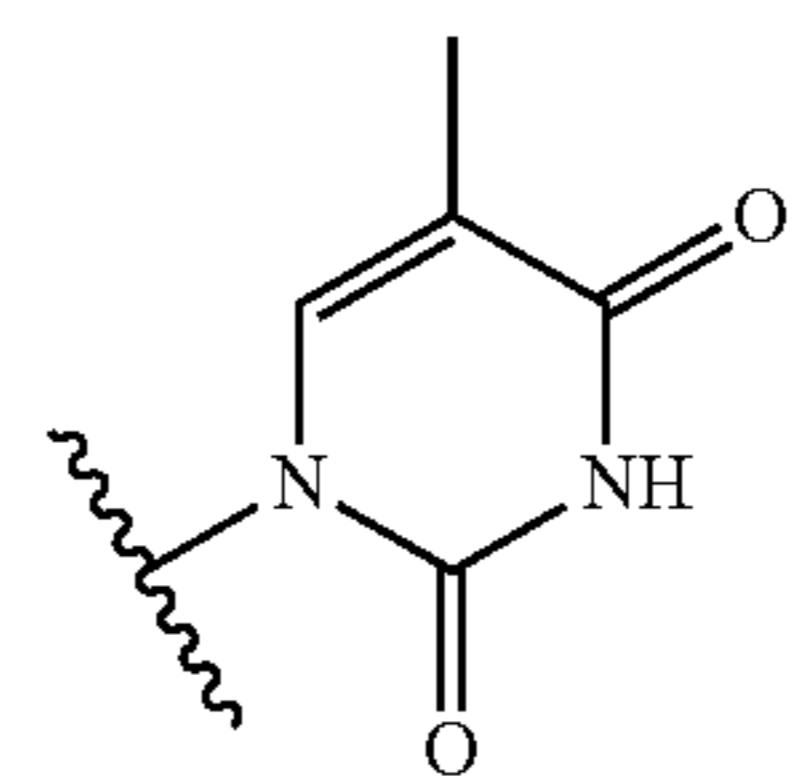


(PC) 45

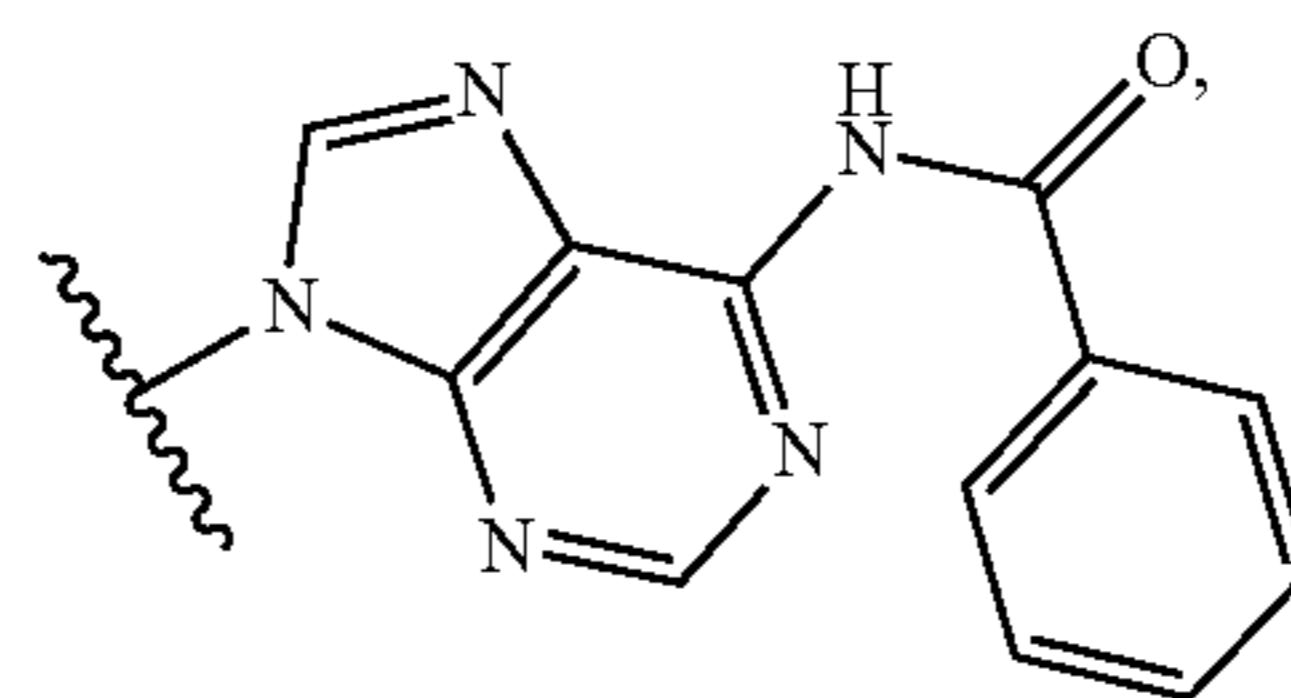


(DPG) 50

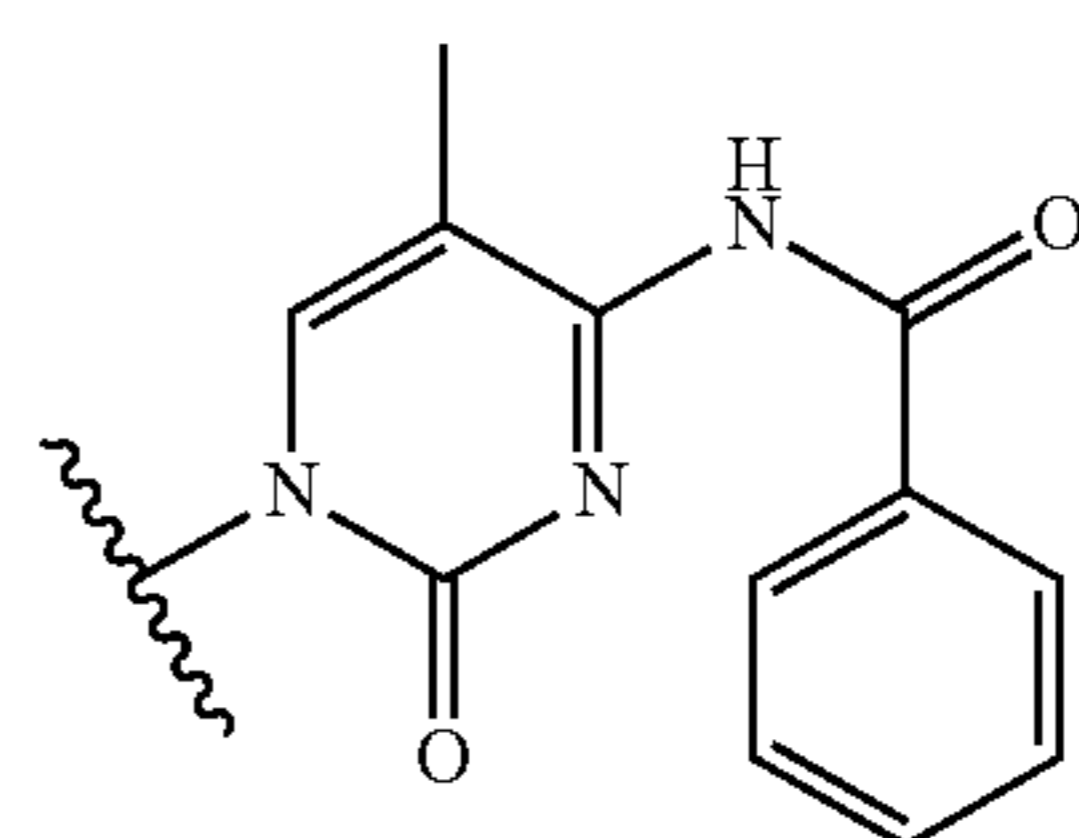
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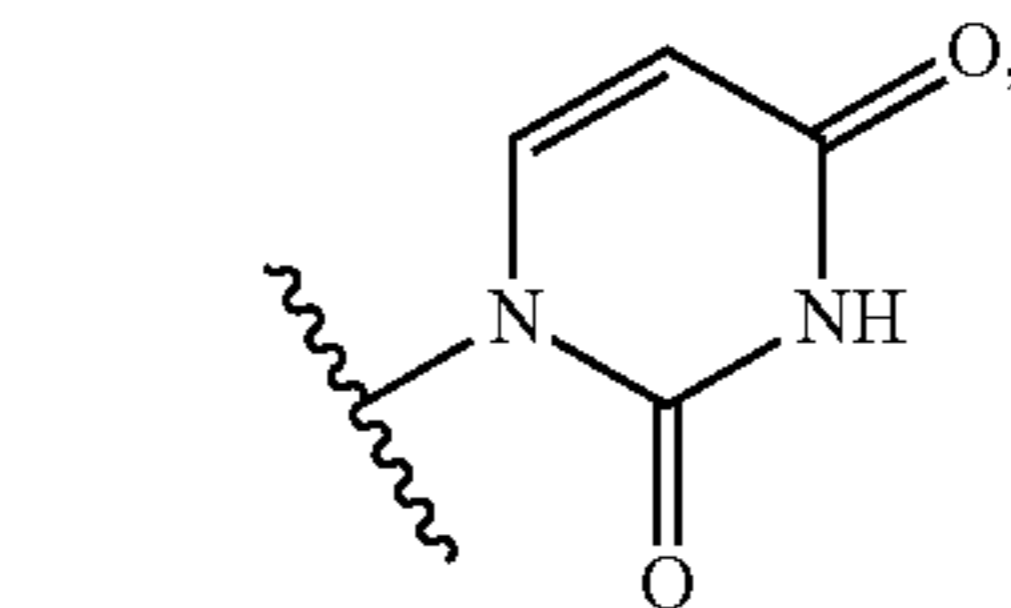
(T) 20



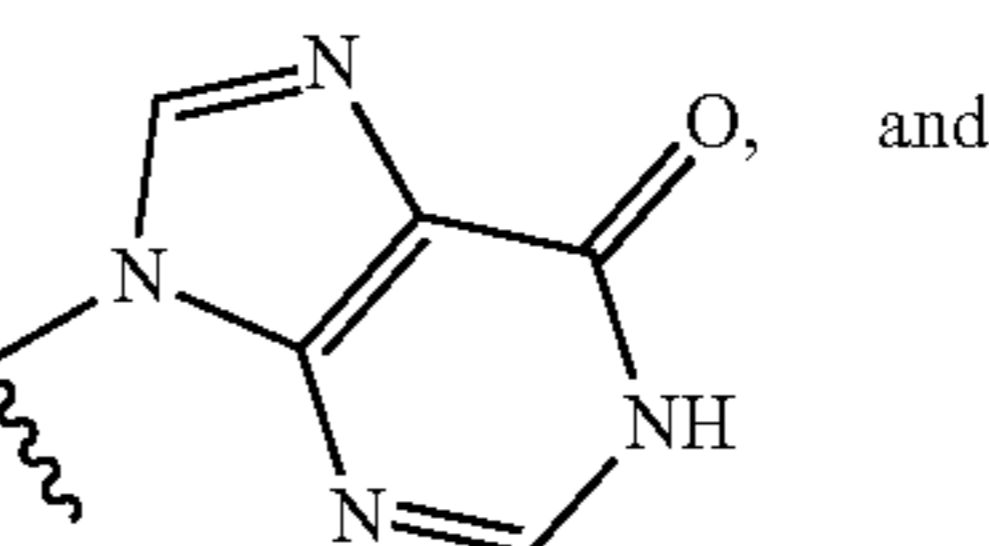
(PA) 30



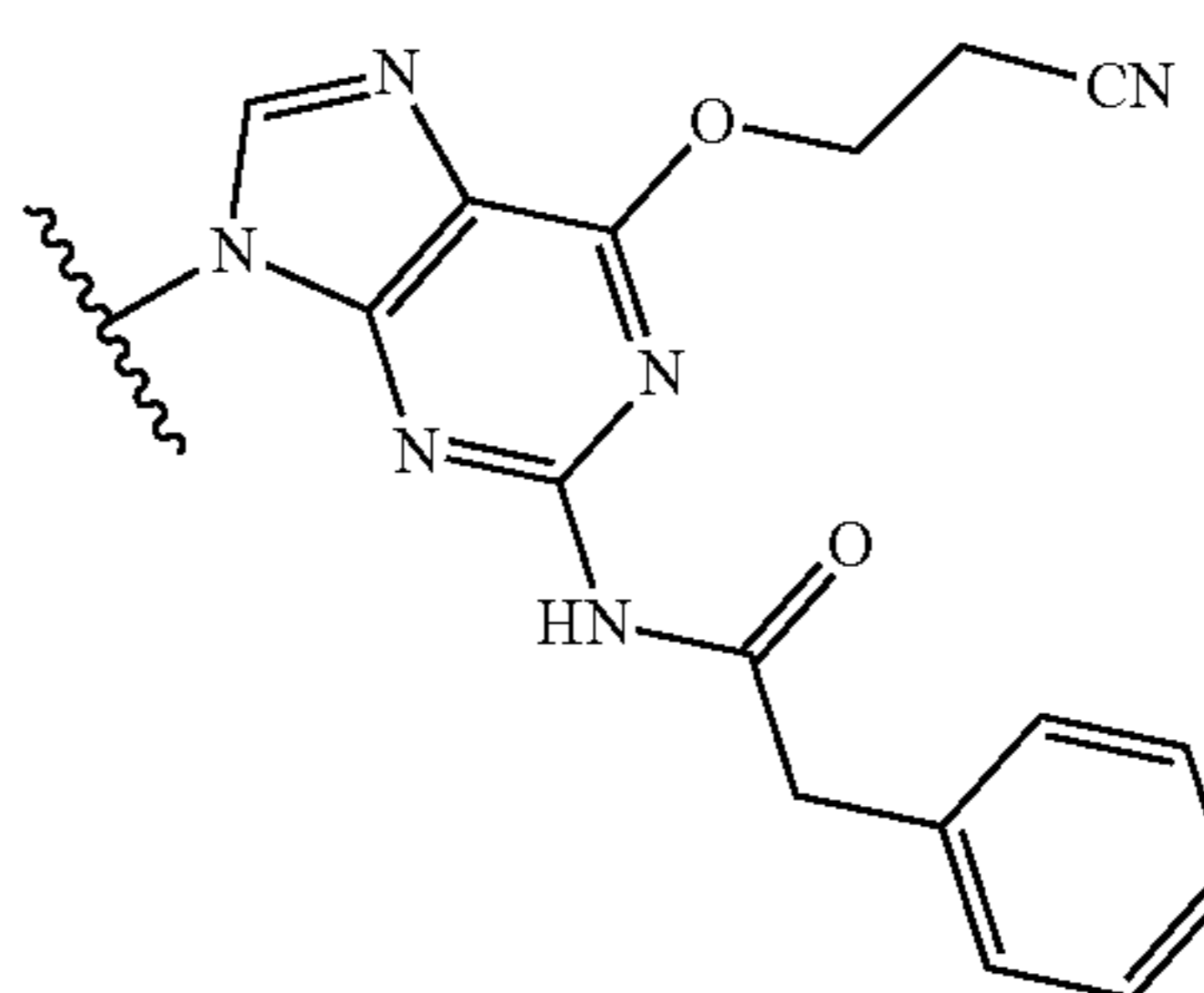
(P5mC) 35



(U) 40



(I) 45



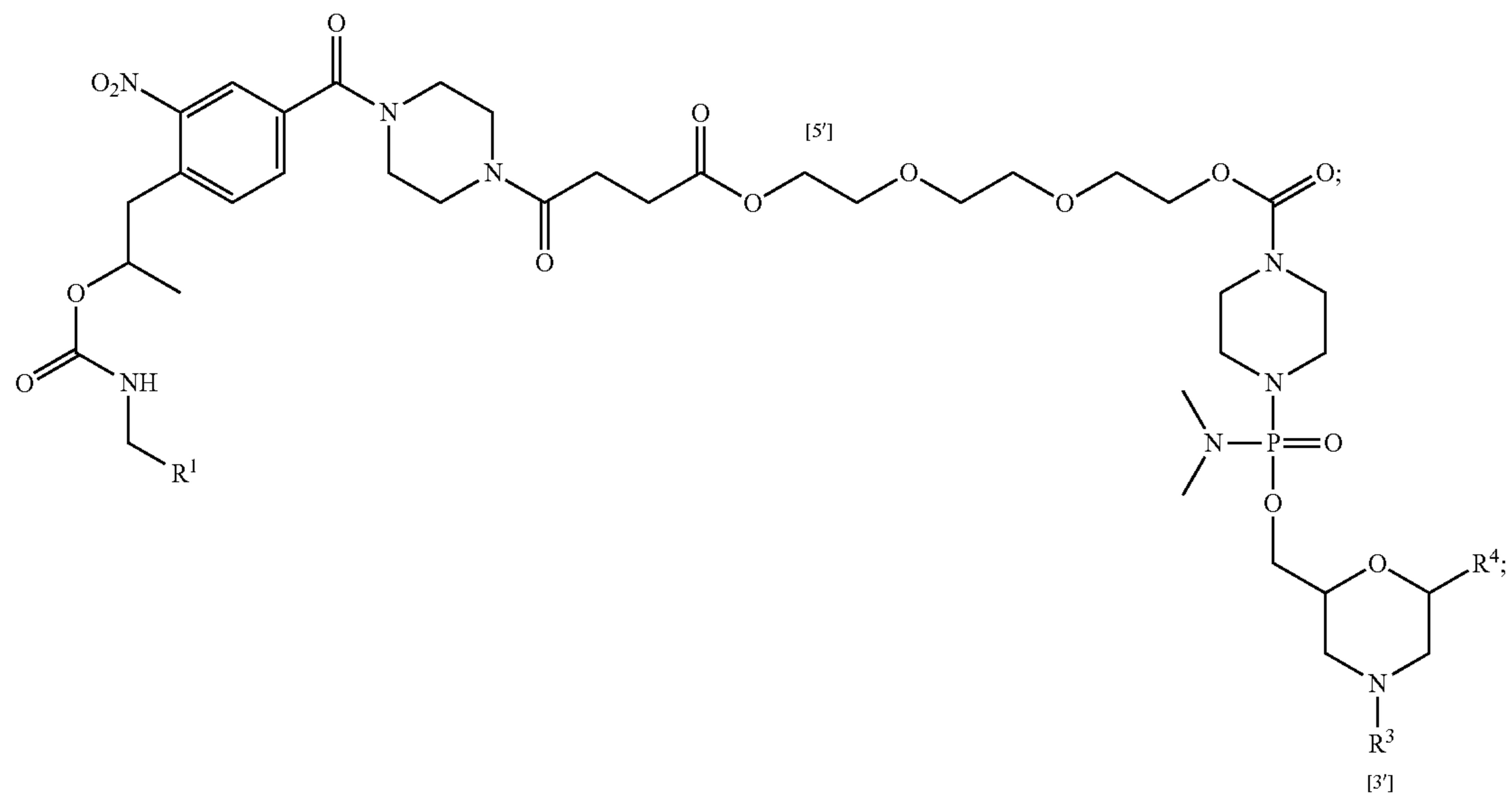
(PG) 55

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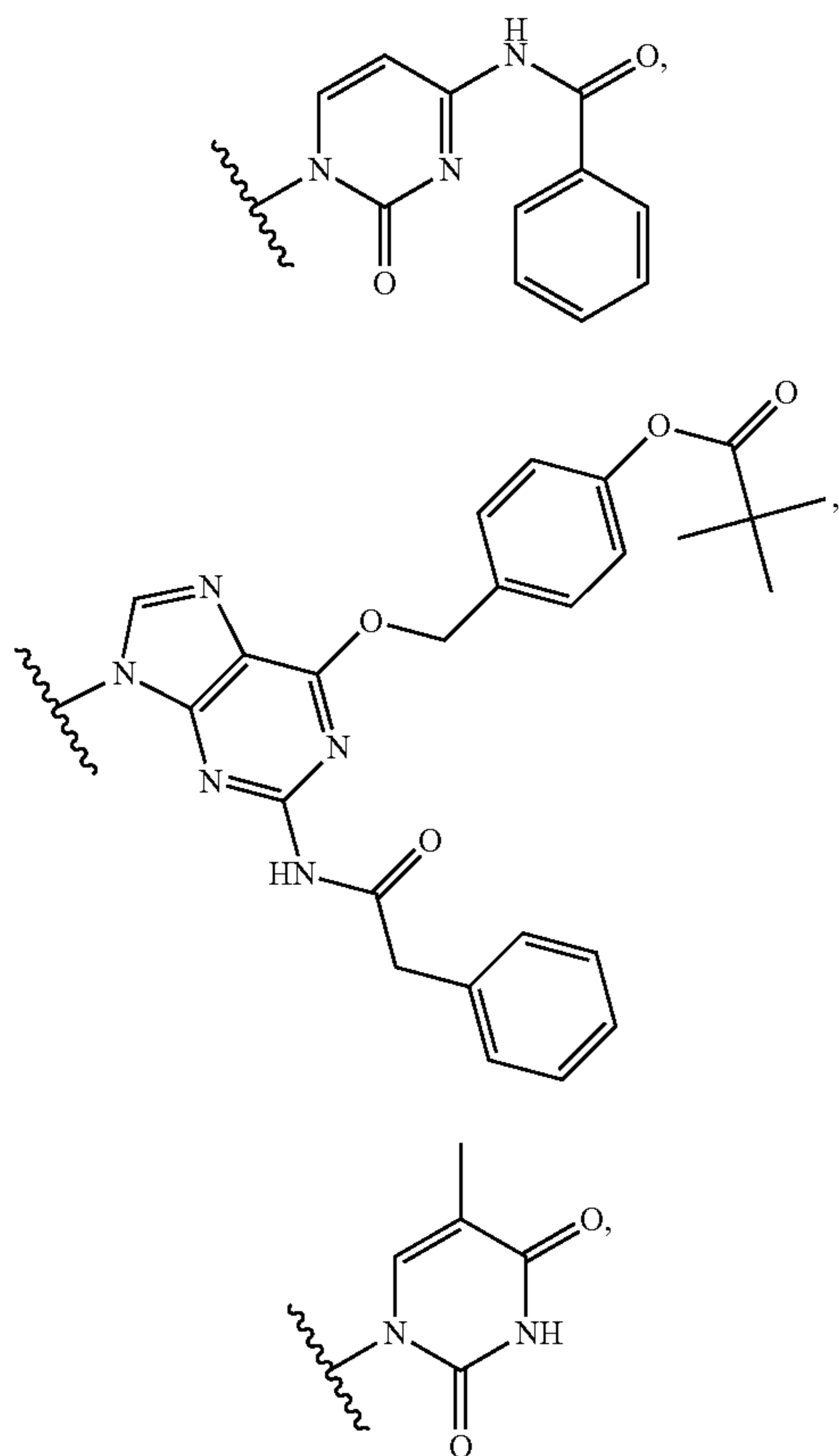
53

to form a compound of Formula (A5):



(A5)

wherein R^1 is a support-medium, R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R^4 is selected from the group consisting of:

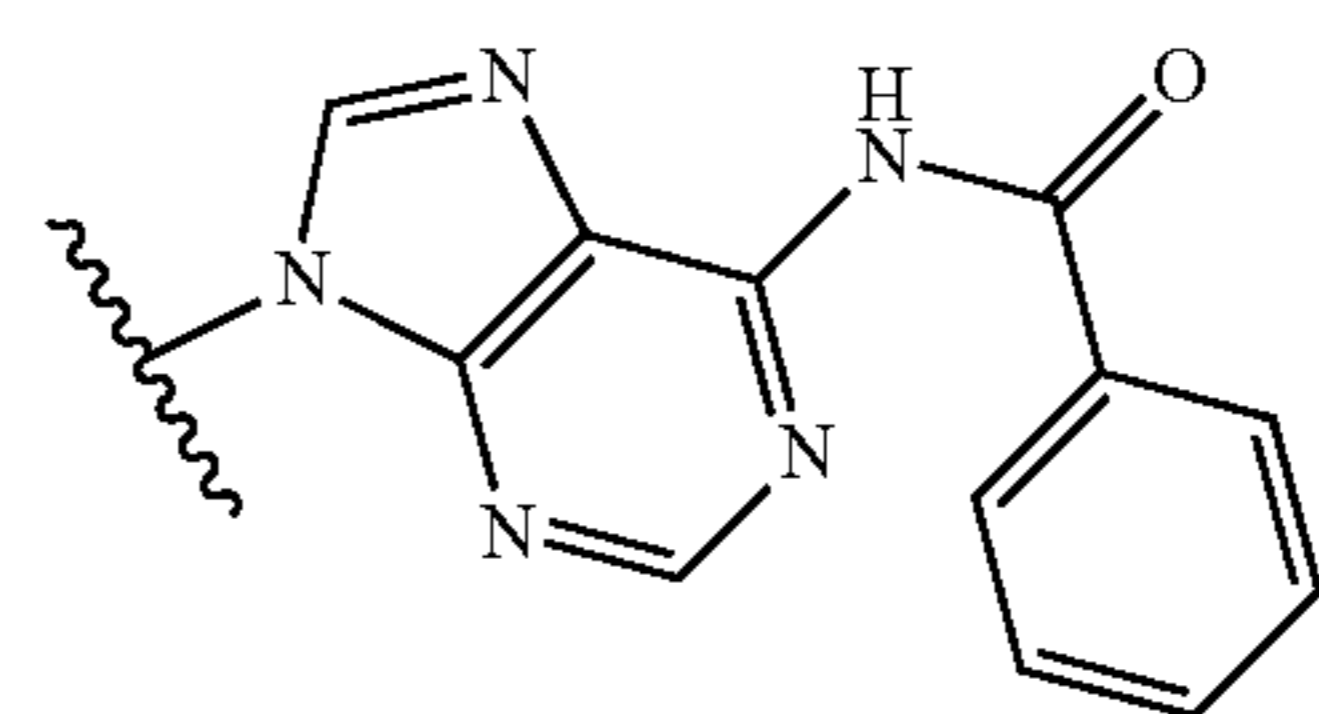


(PC)

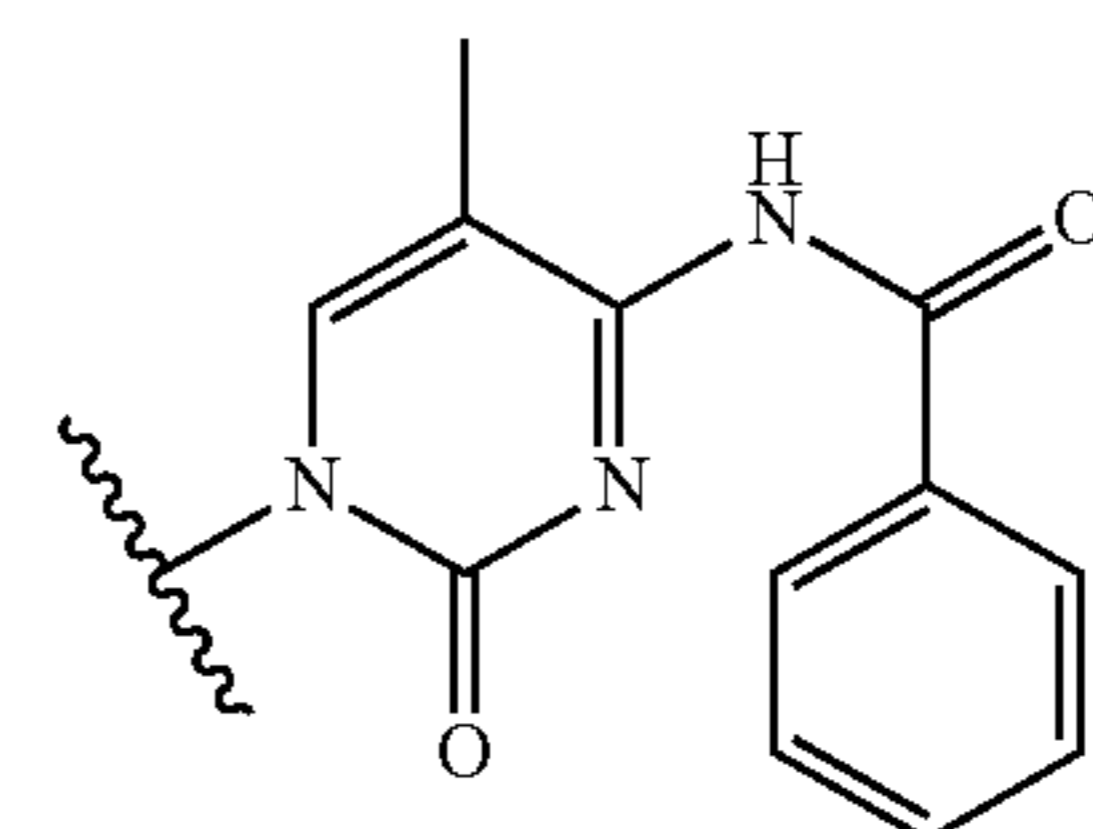
(DPG)

(T)

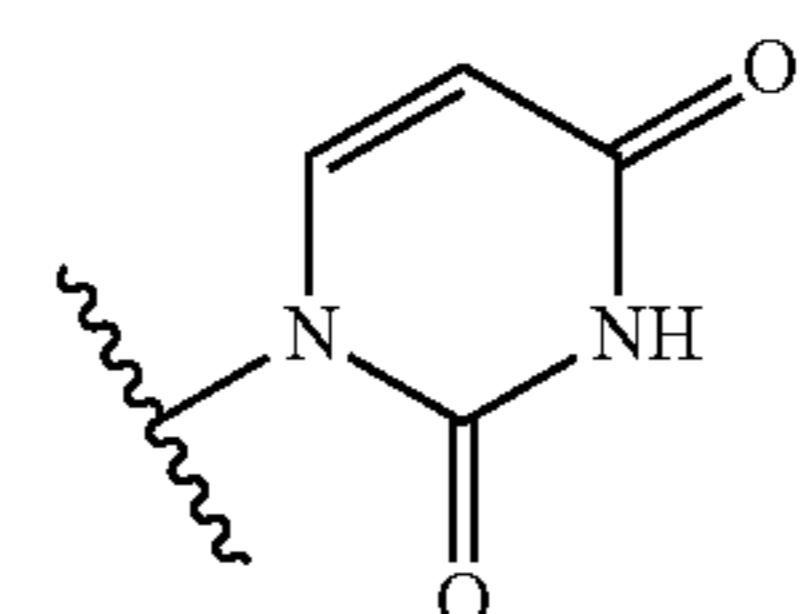
-continued



(PA)



(P5mC)



(U)



(I)

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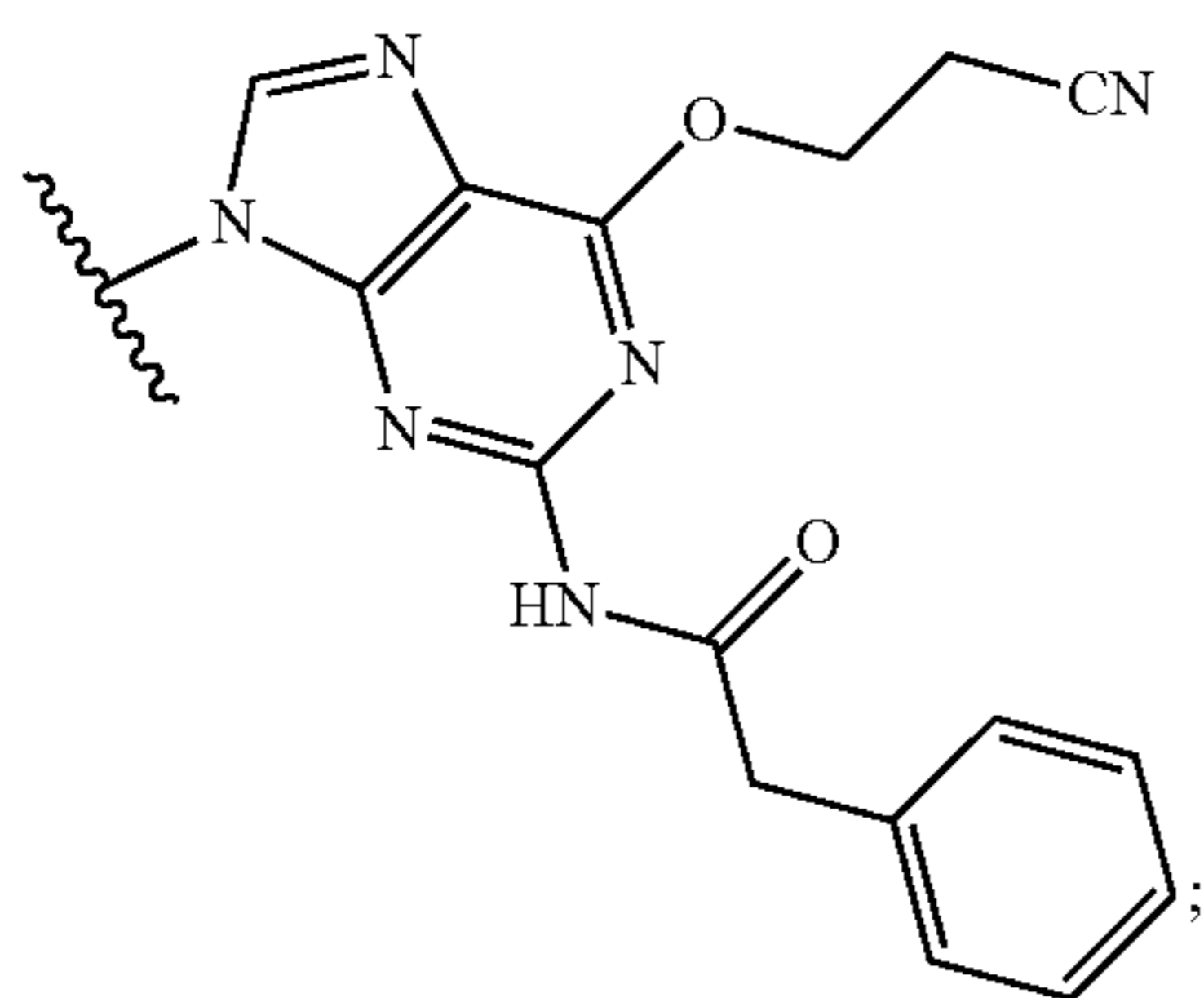
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(PG)

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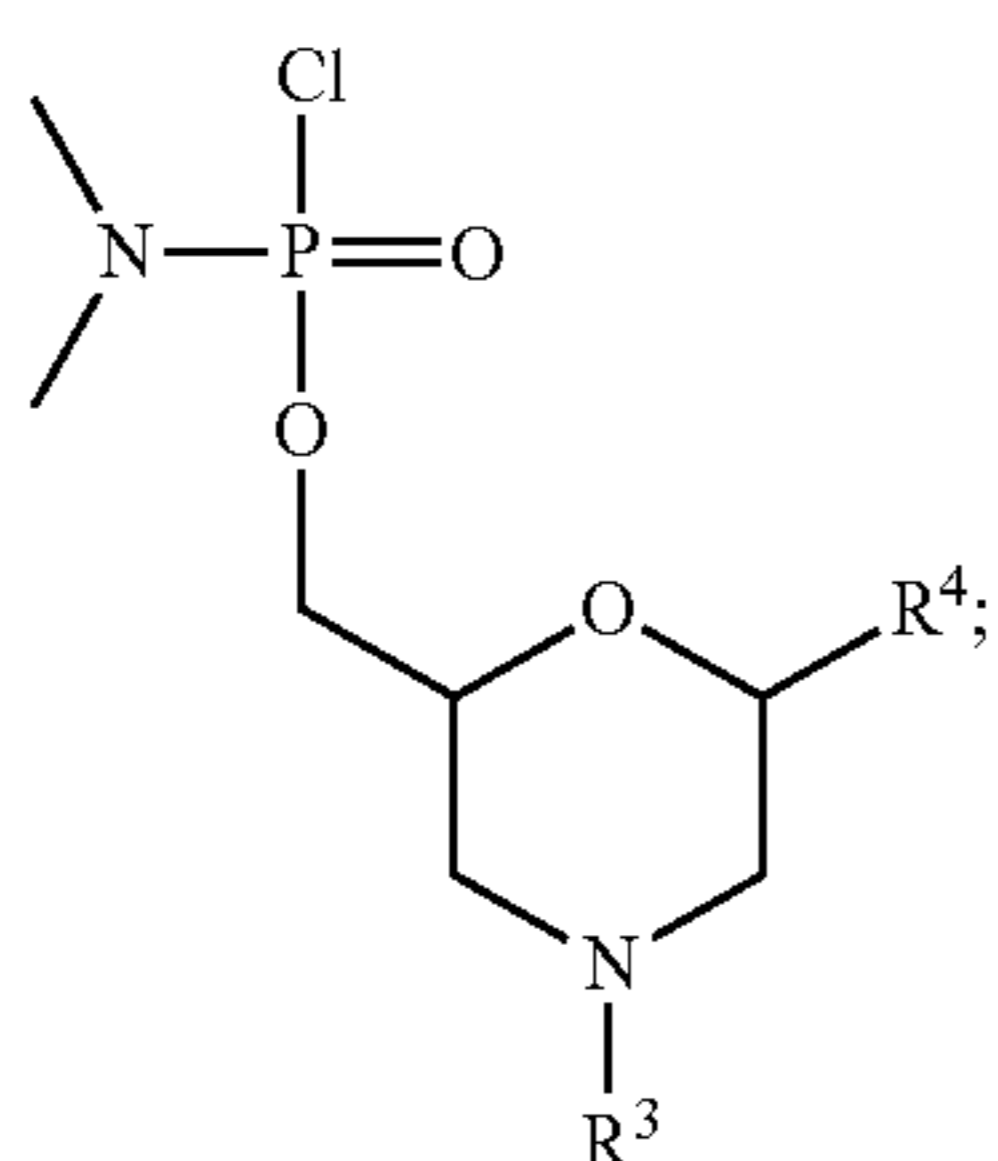
10

15

(e) performing n-1 iterations of the sequential steps of:

(e1) contacting the product formed by the immediately prior step with a deblocking agent; and

(e2) contacting the compound formed by the immediately prior step with a compound of Formula (A8):



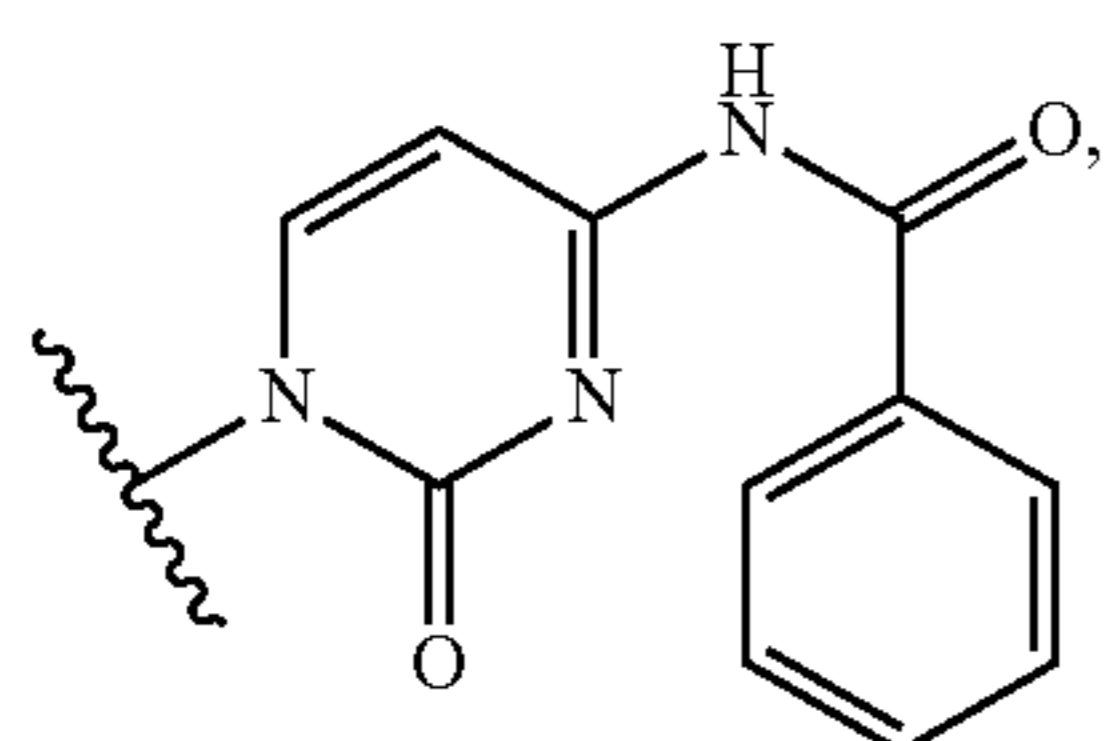
(A8)

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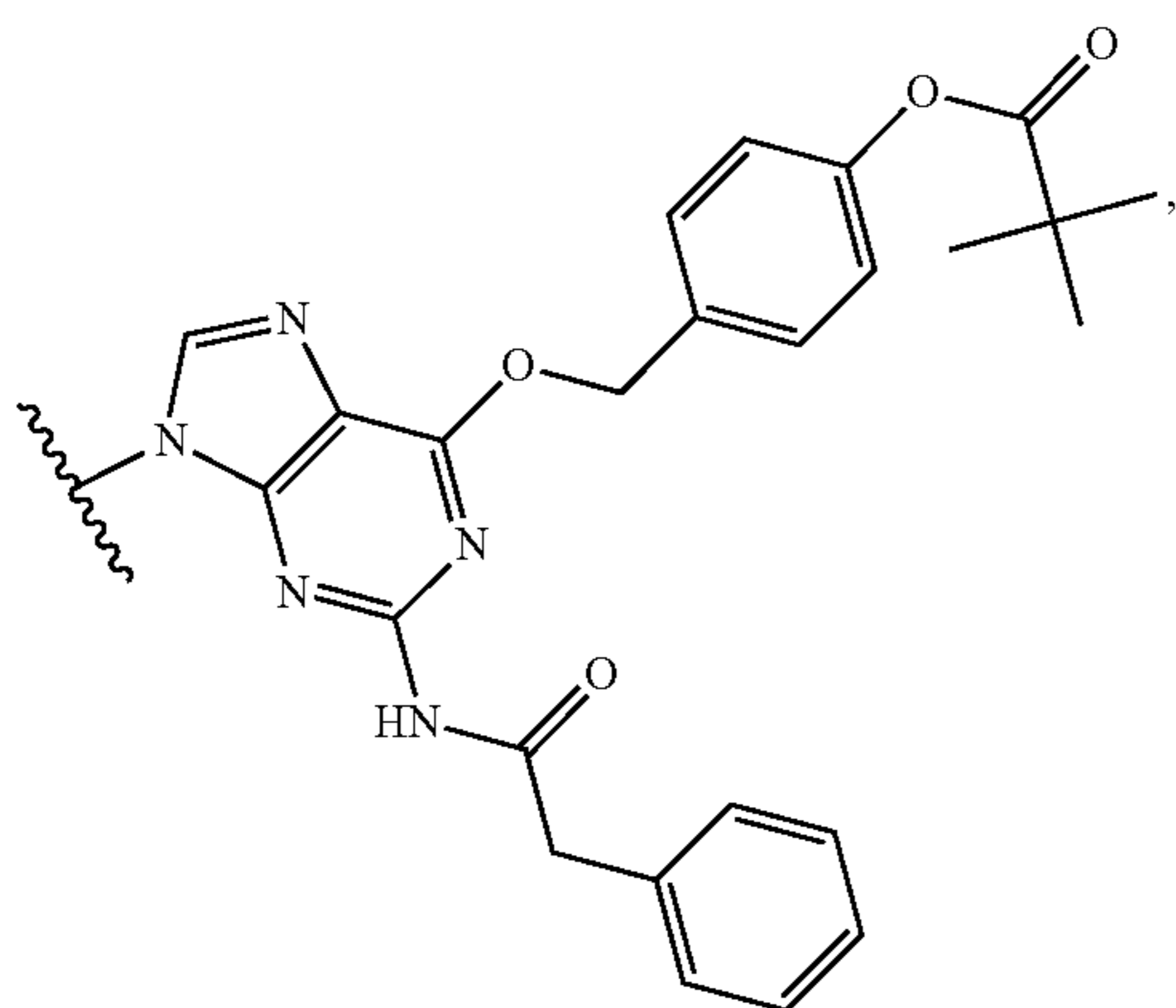
wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each compound of Formula (A8), selected from the group consisting of:



(PC)

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(DPG)

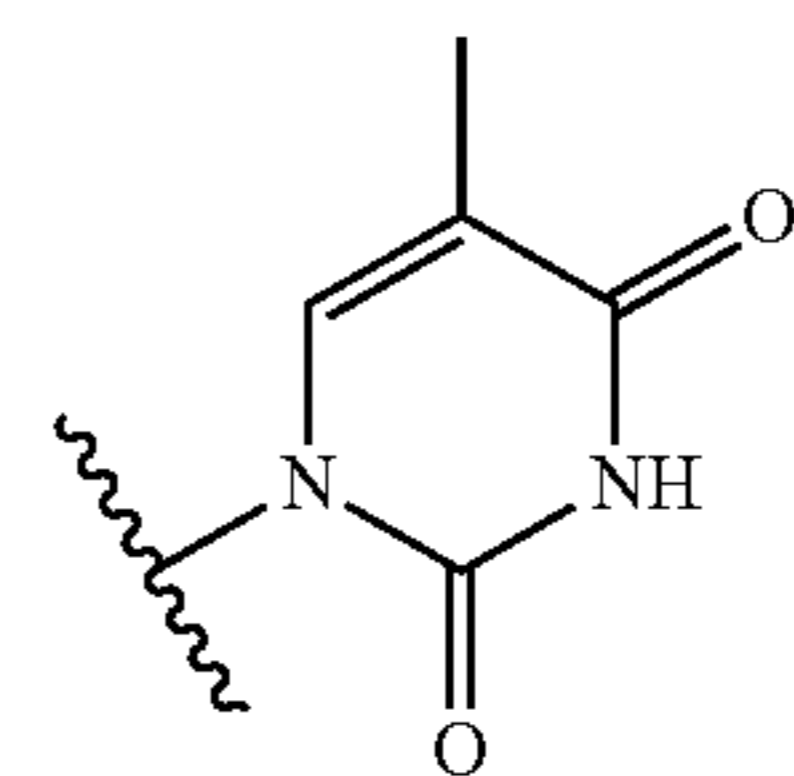
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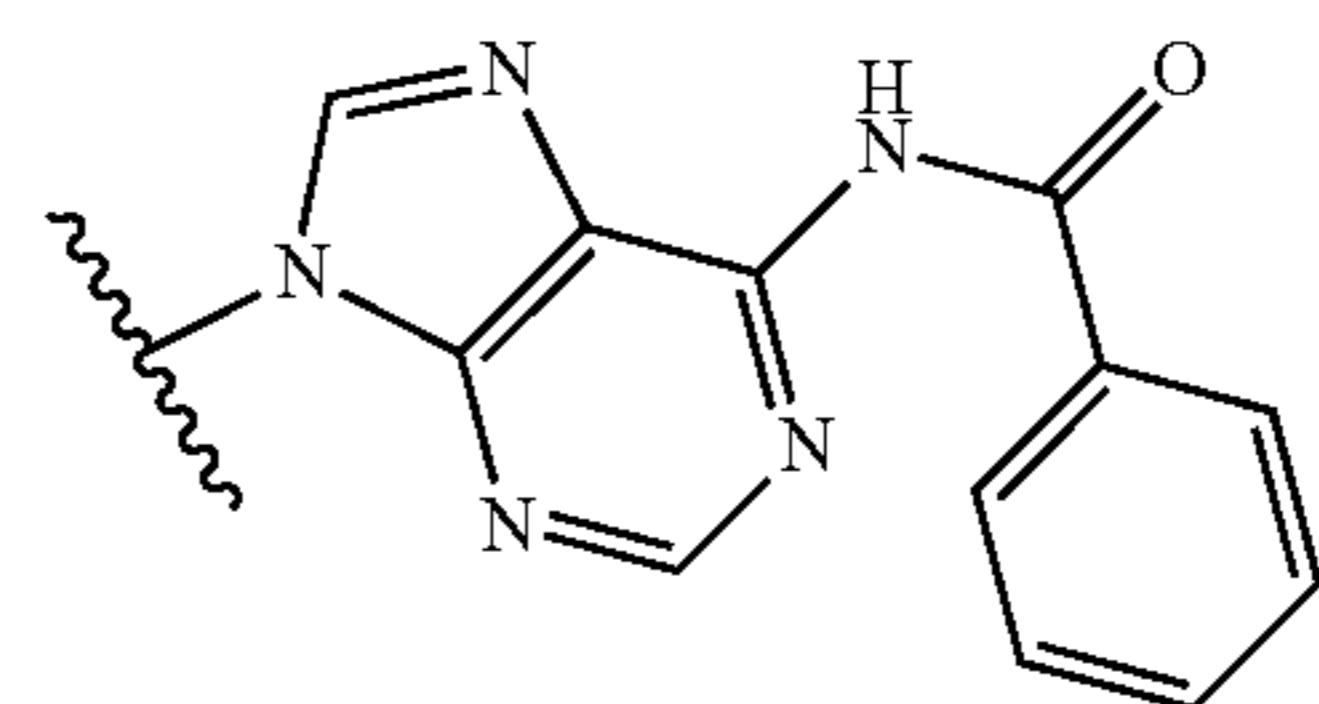
65

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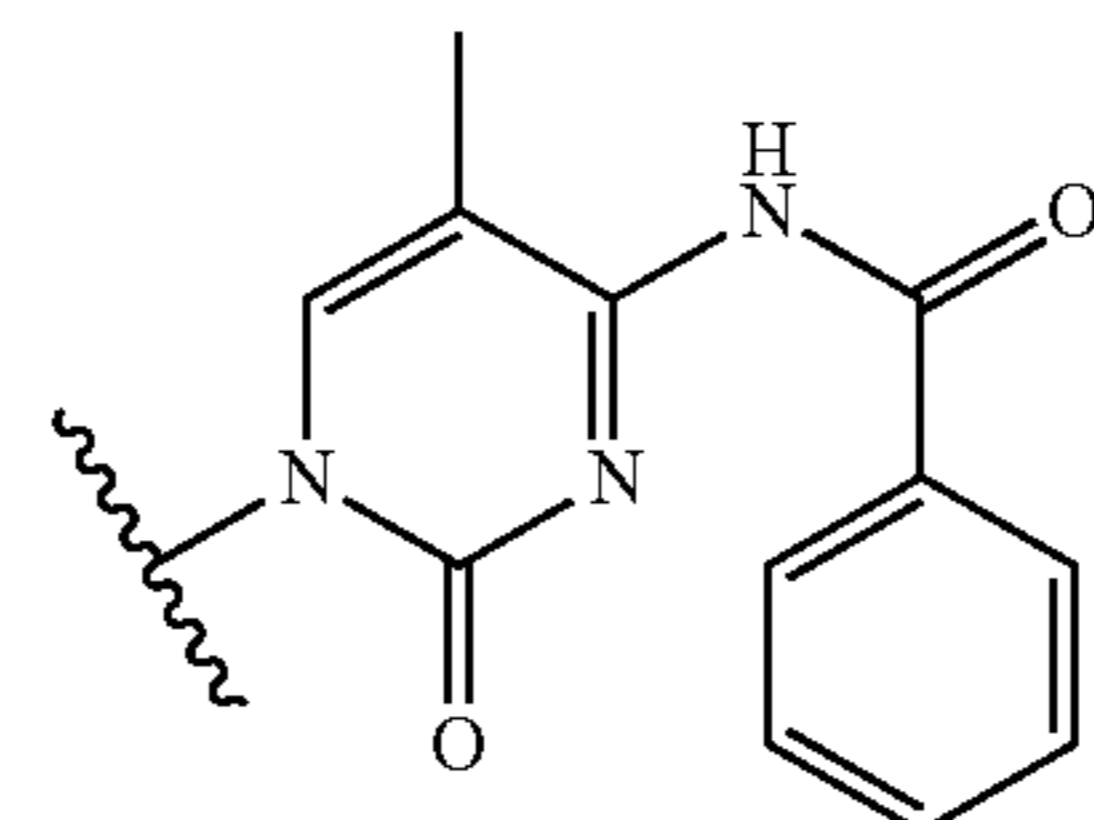
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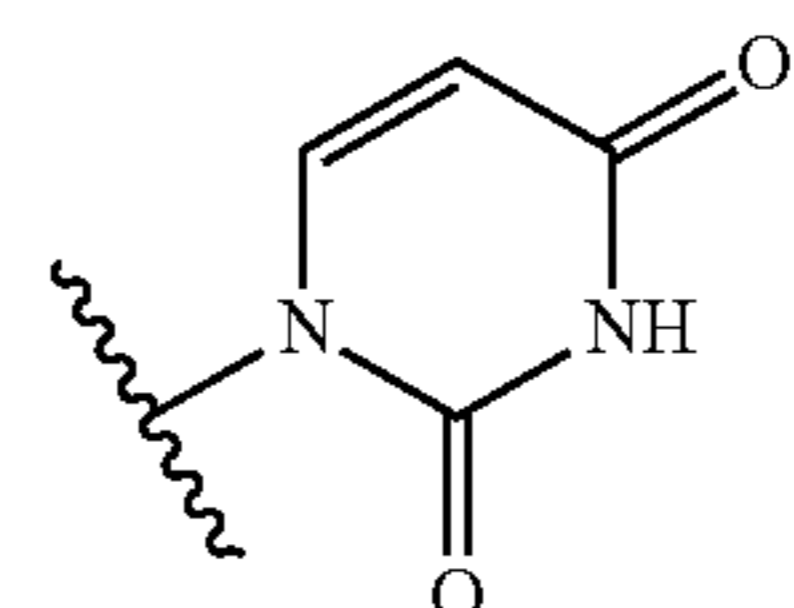
(T)



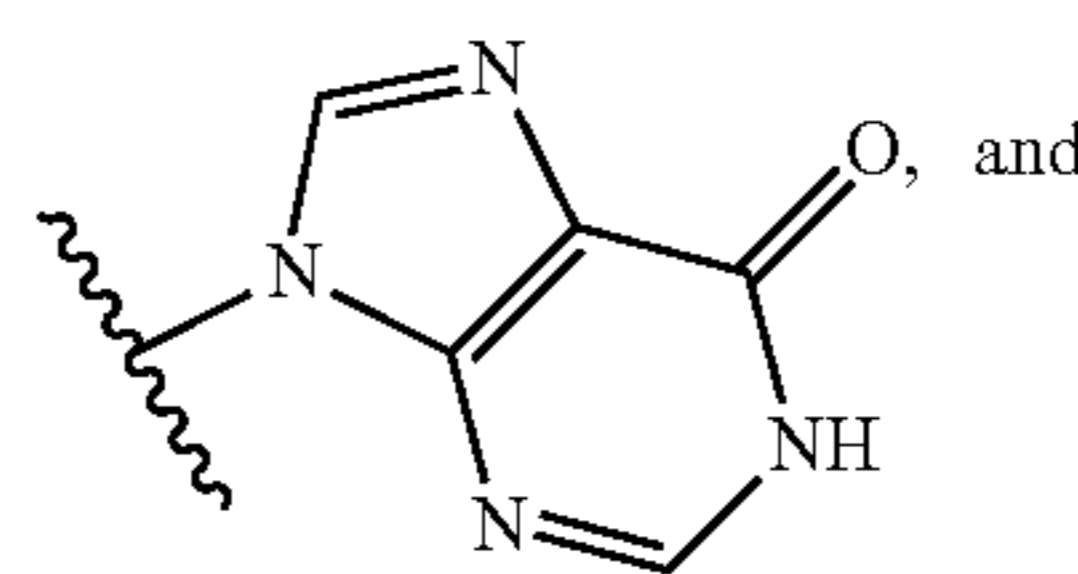
(PA)



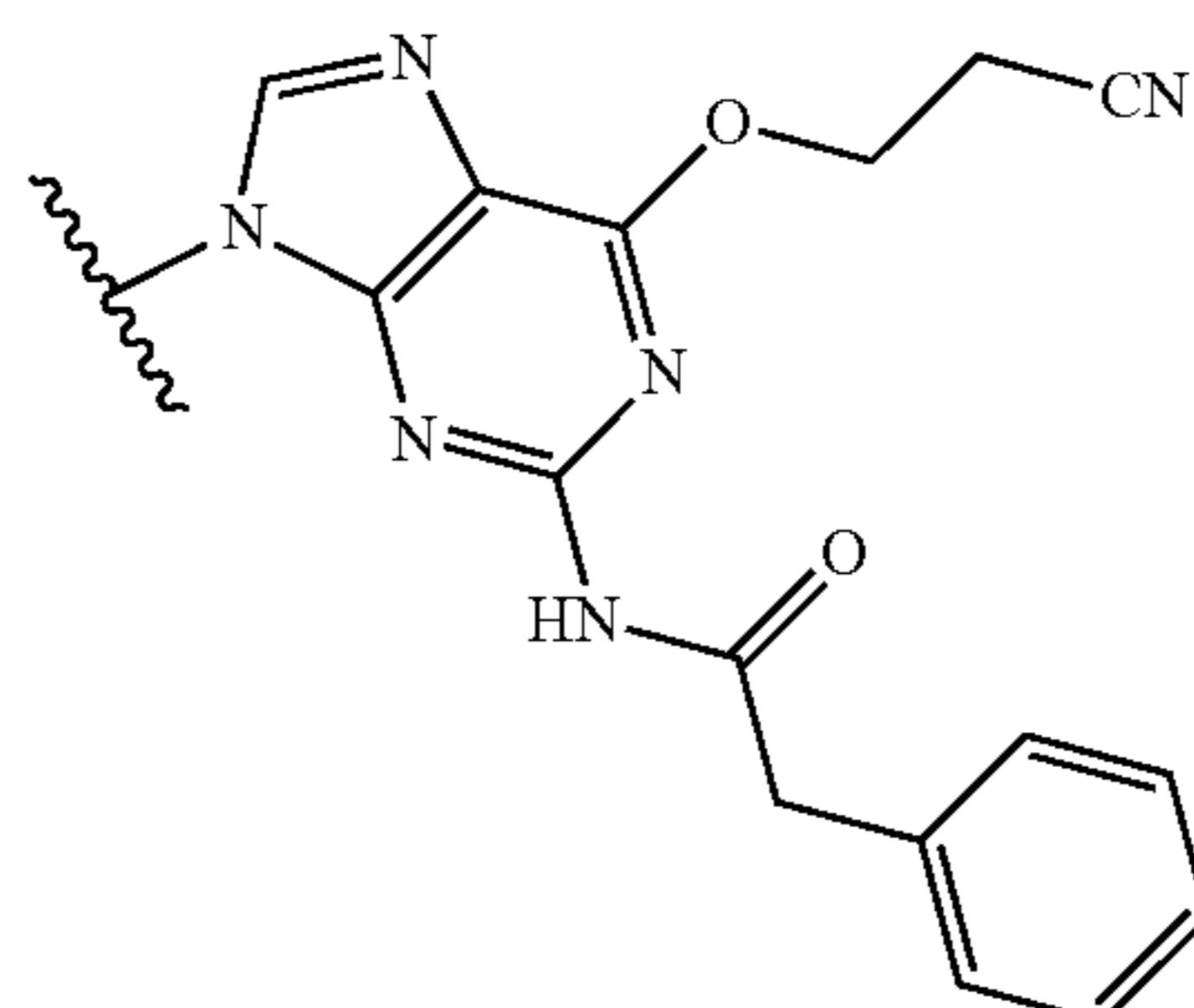
(P5mC)



(U)



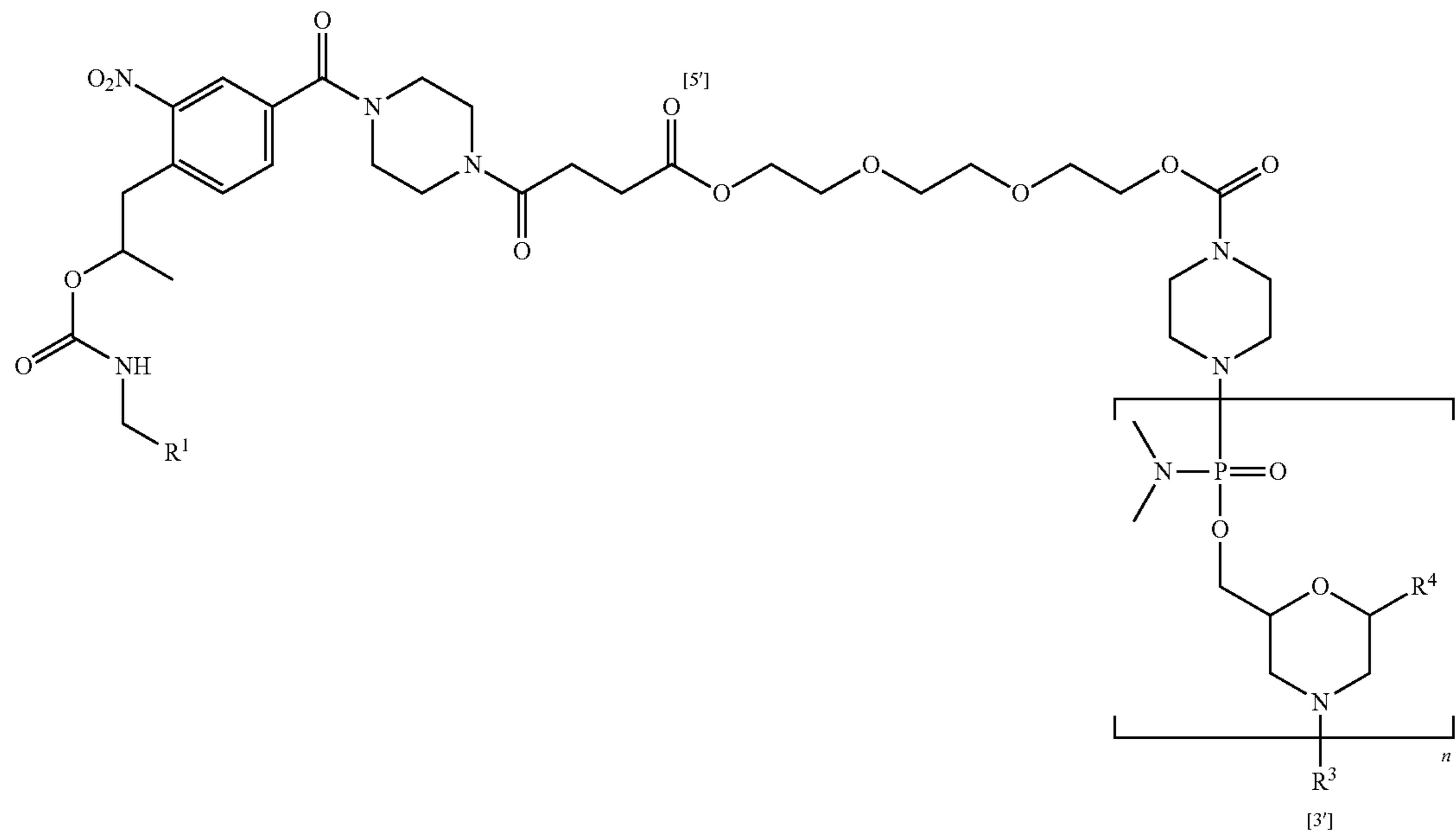
(I)



(PG)

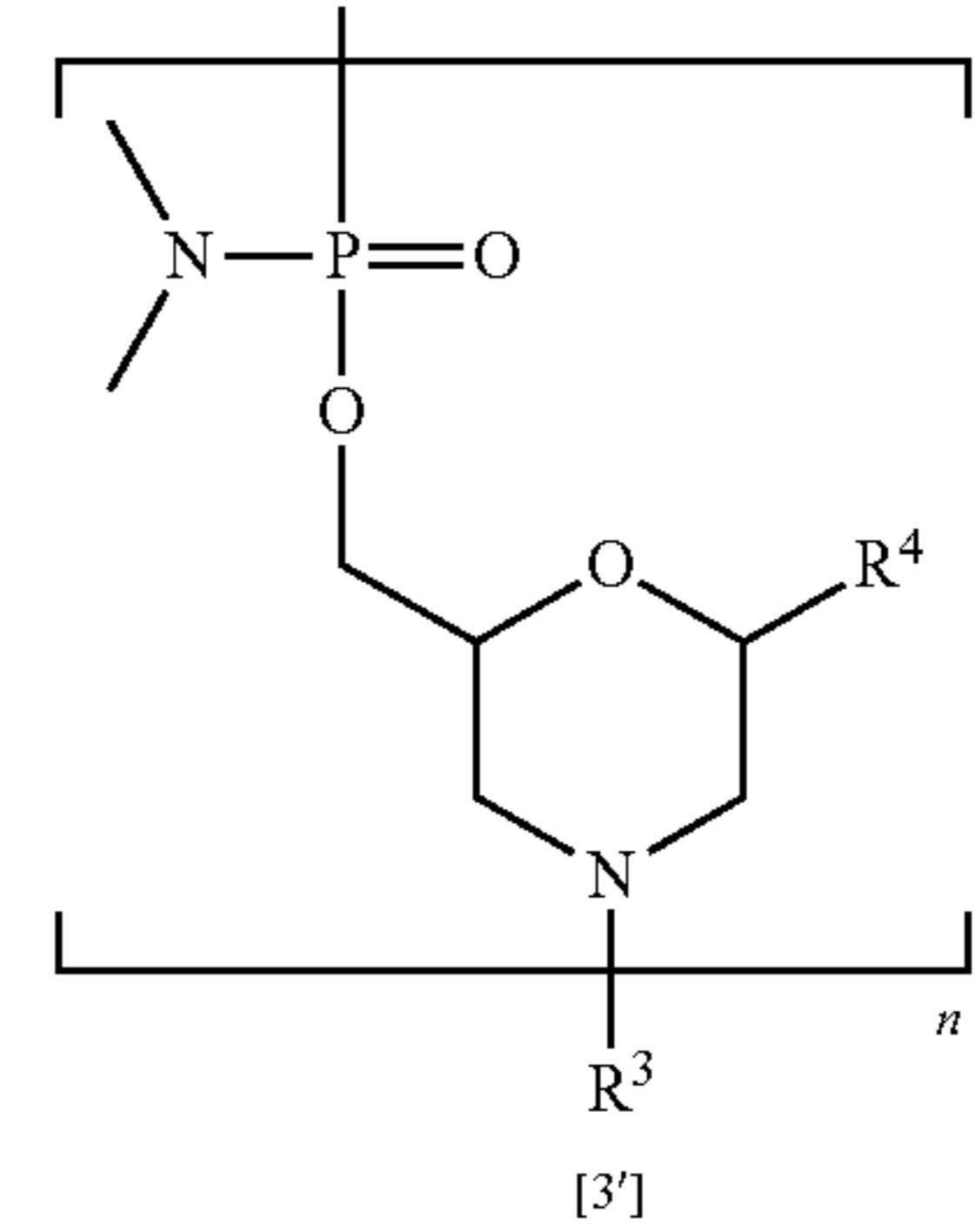
57

to form a compound of Formula (A9):



58

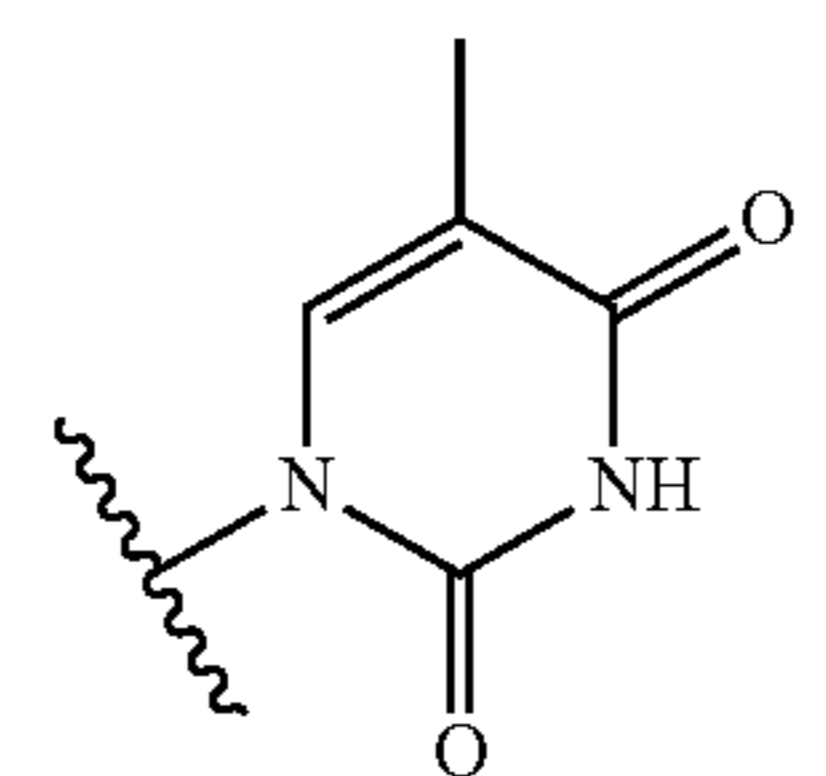
(A9)



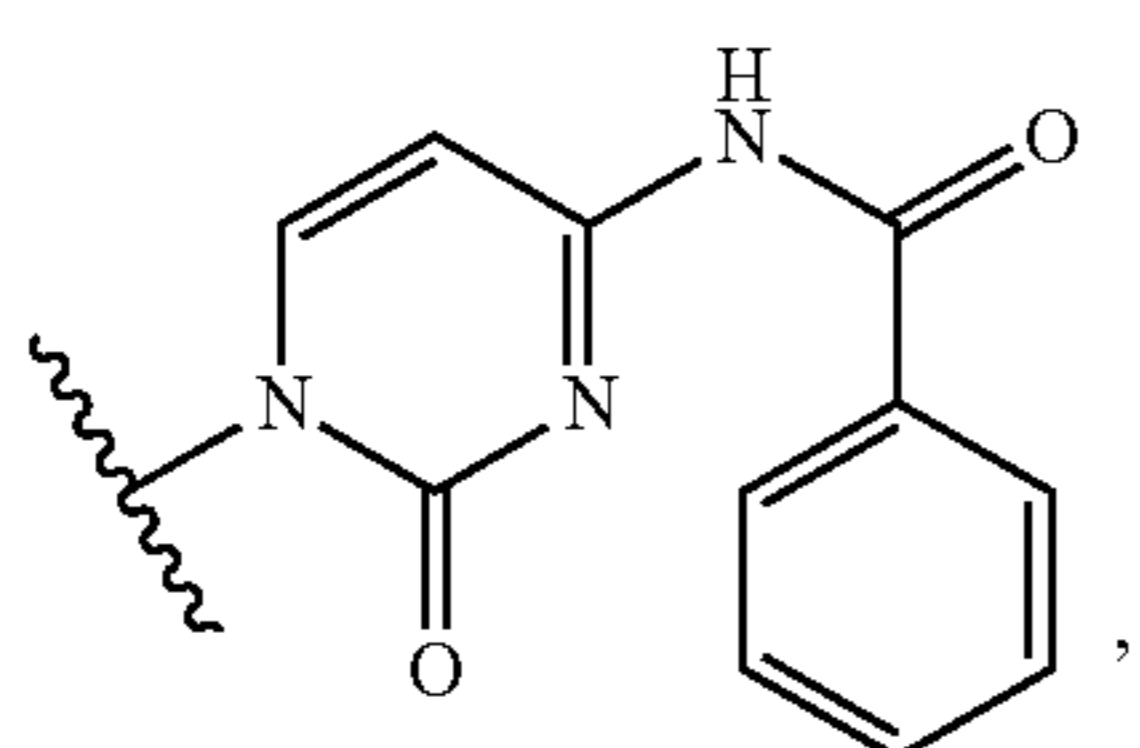
wherein *n* is an integer from 10 to 40, R¹ is a support-medium, R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each occurrence, selected from the group consisting of:

-continued

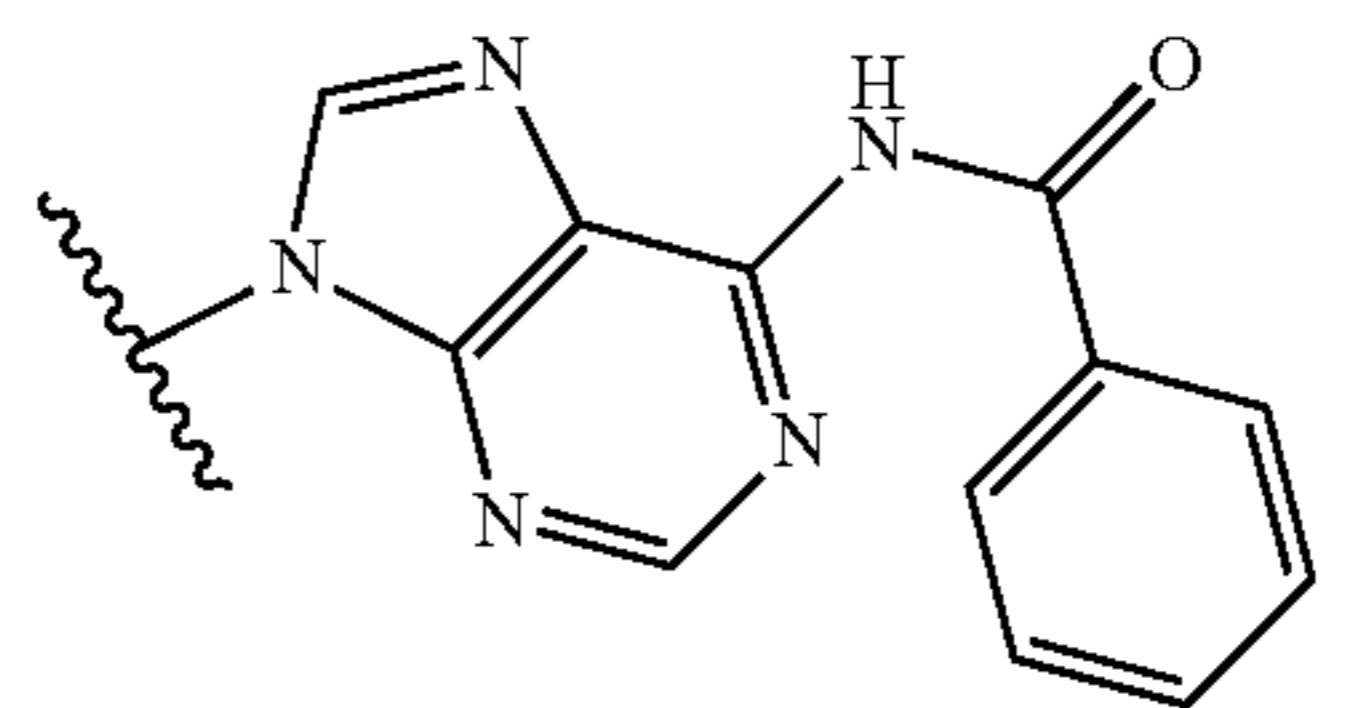
(T)



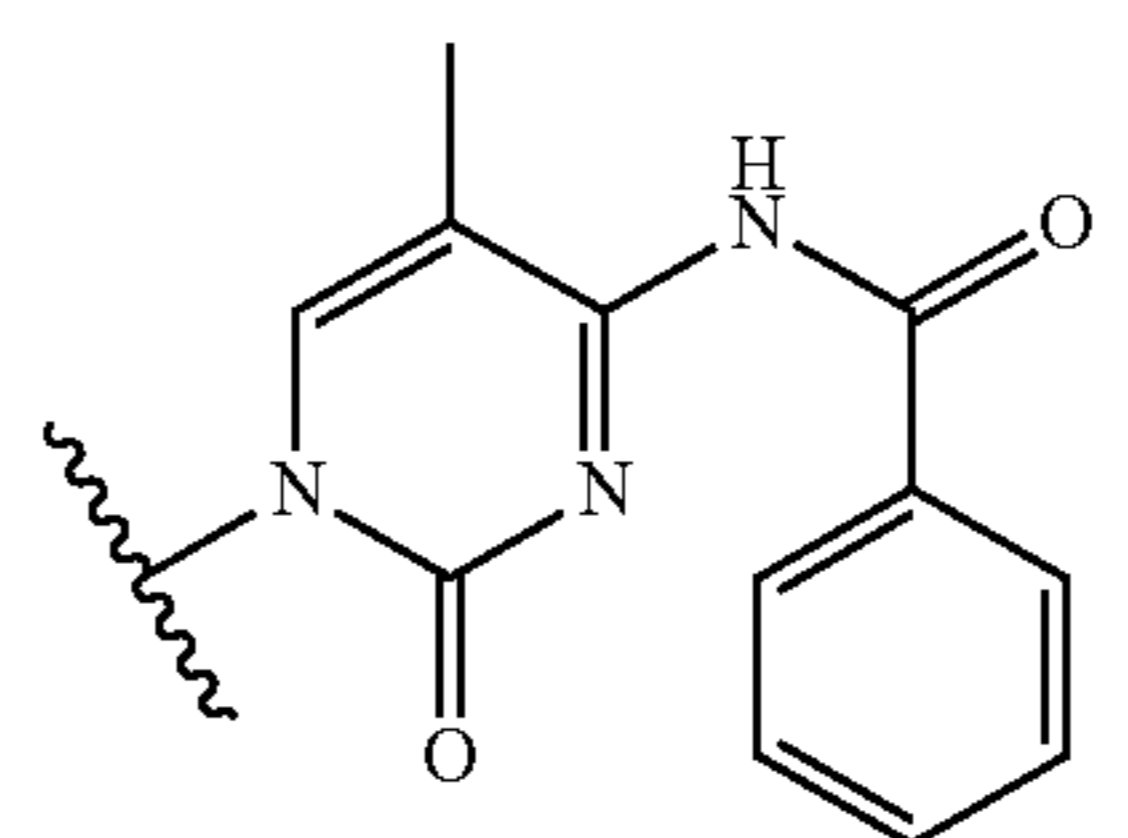
(PC)



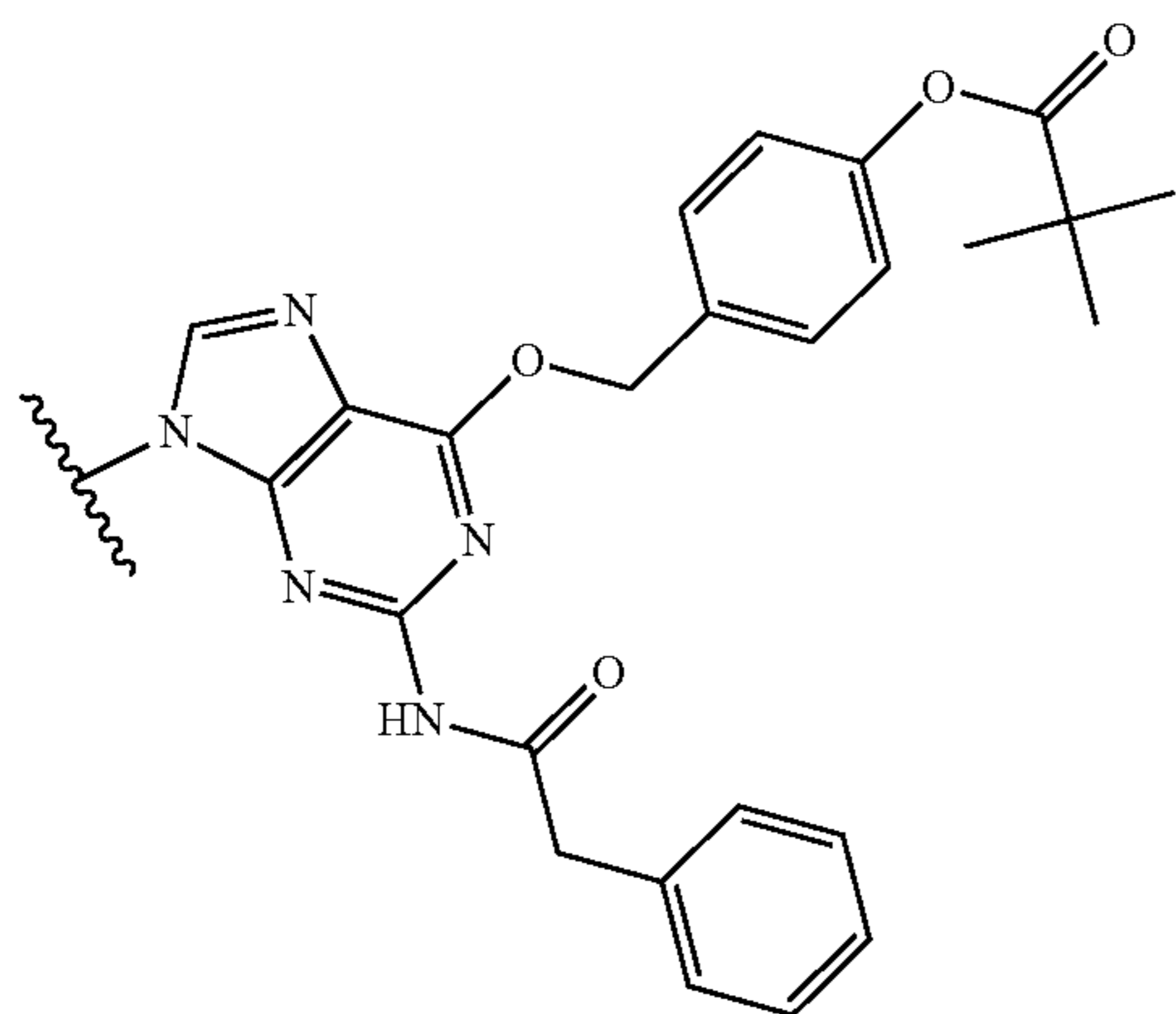
(PA)



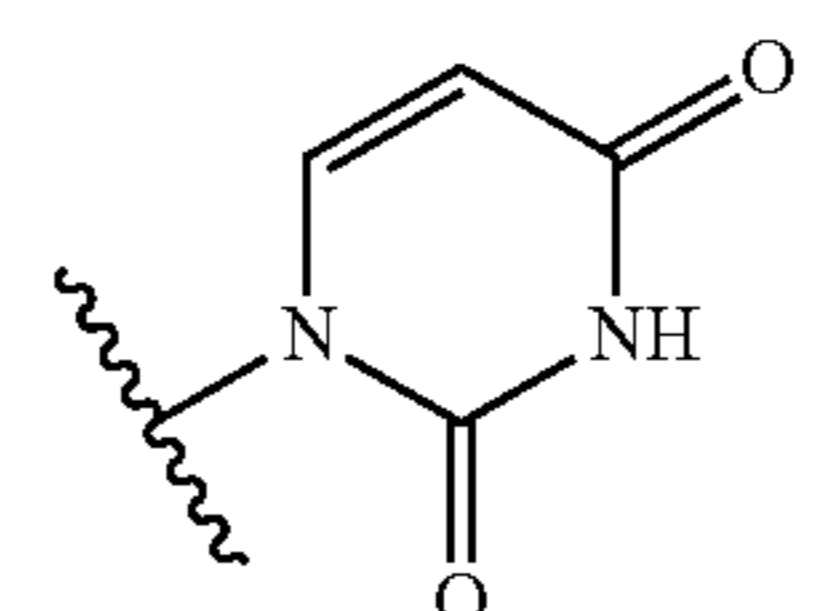
(P5mC)



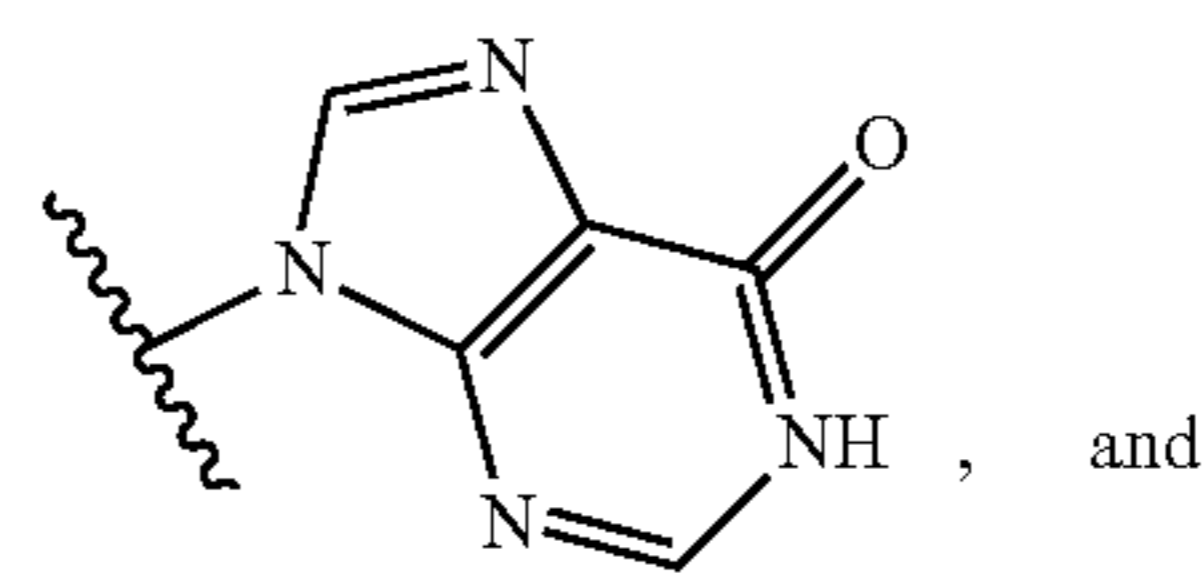
(DPG)



(U)



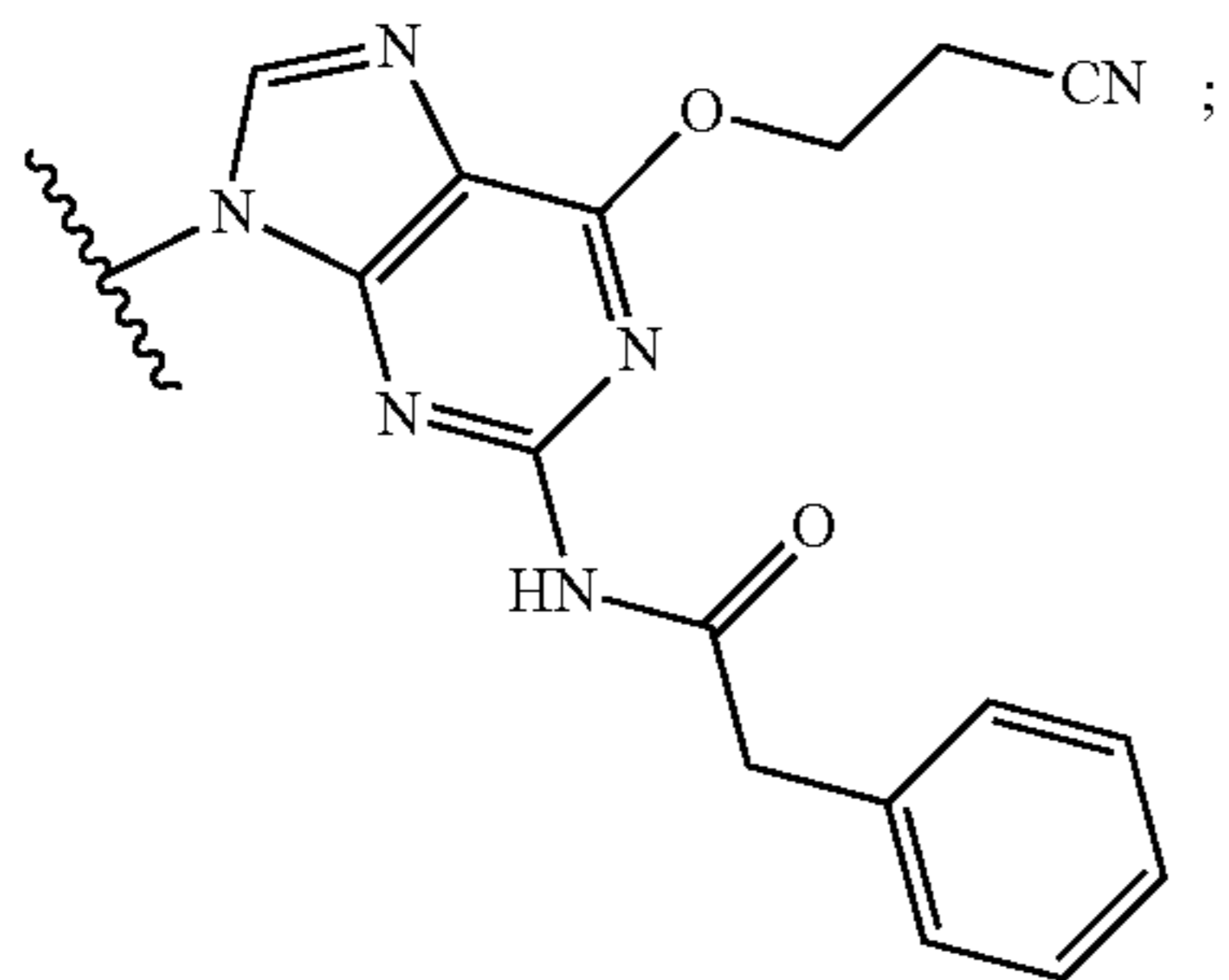
(I)



and

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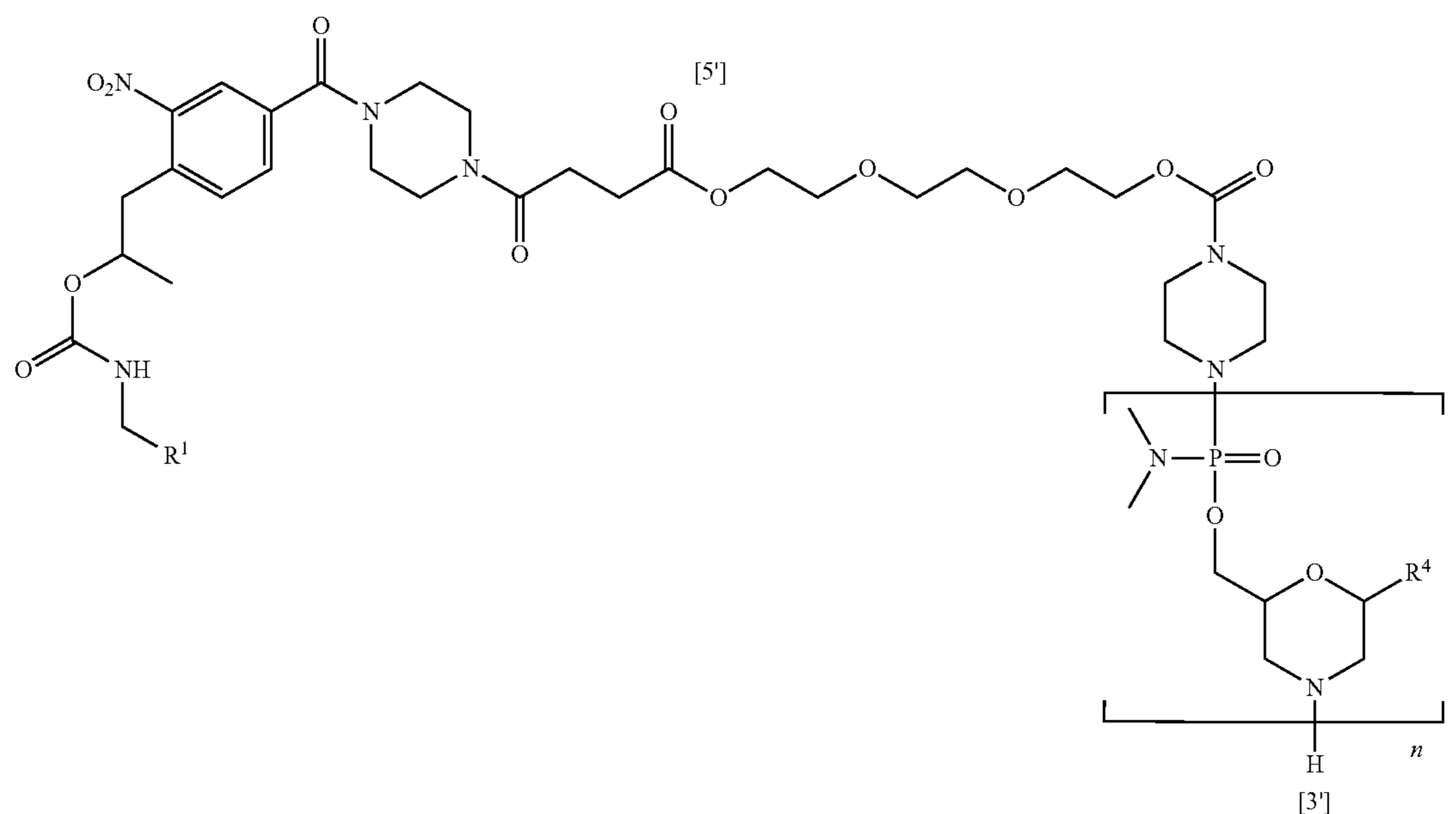


(PG)

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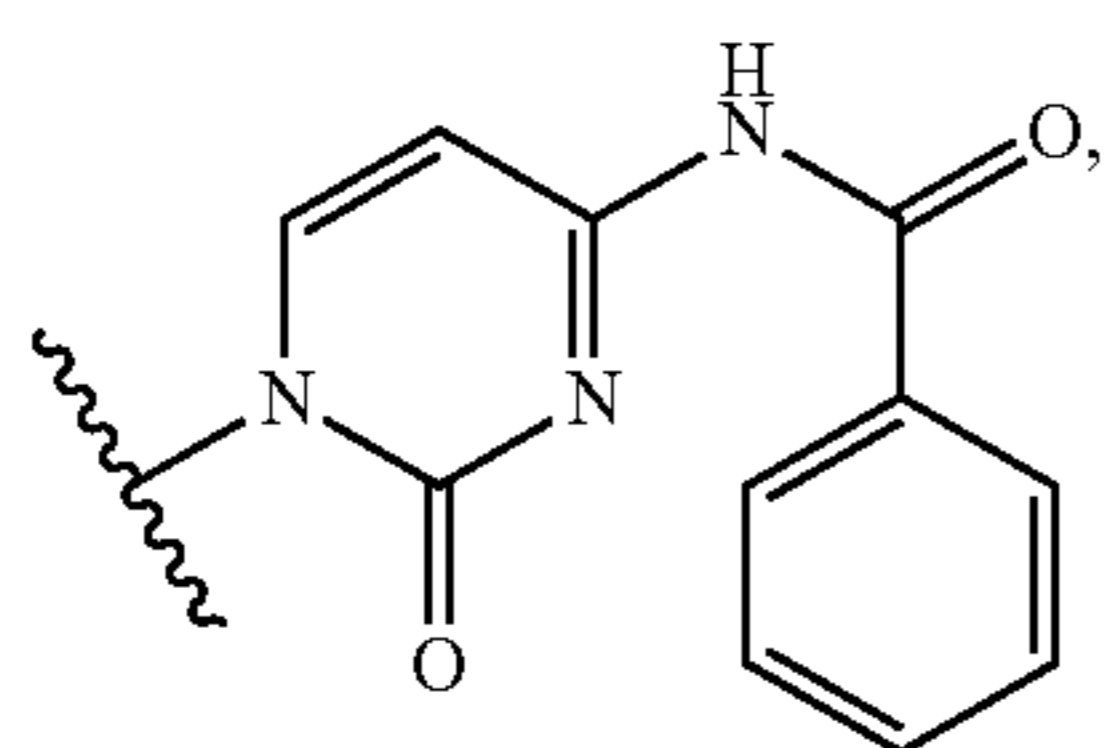
10

(f) contacting the compound of Formula (A9) with a ¹⁵ deblocking agent to form a compound of Formula (A10).



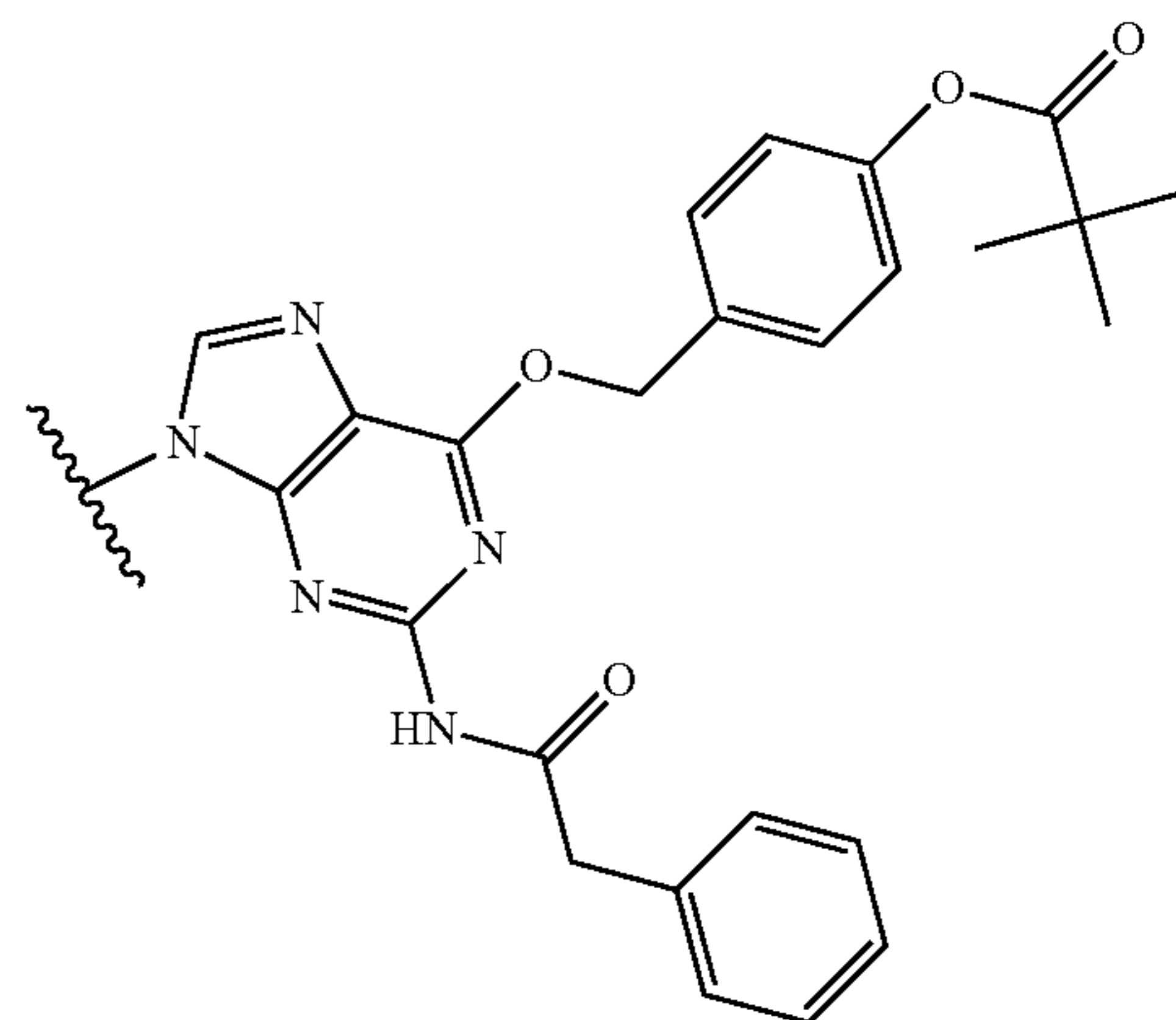
(A10)

wherein n is an integer from 10 to 40, R¹ is a support-medium, and R⁴ is, independently for each occurrence, selected from the group consisting of:



-continued

(DPG)



(PC)

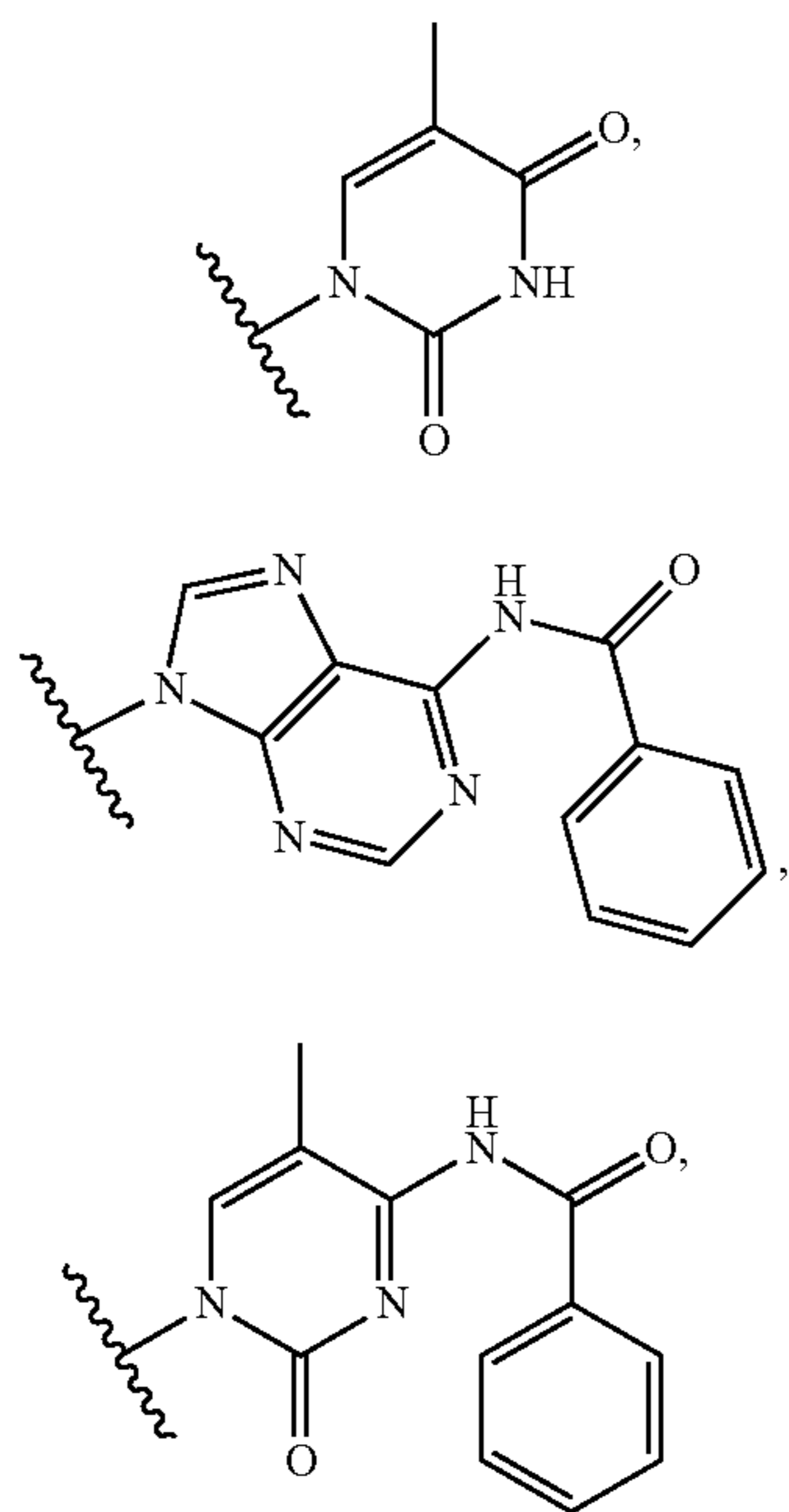
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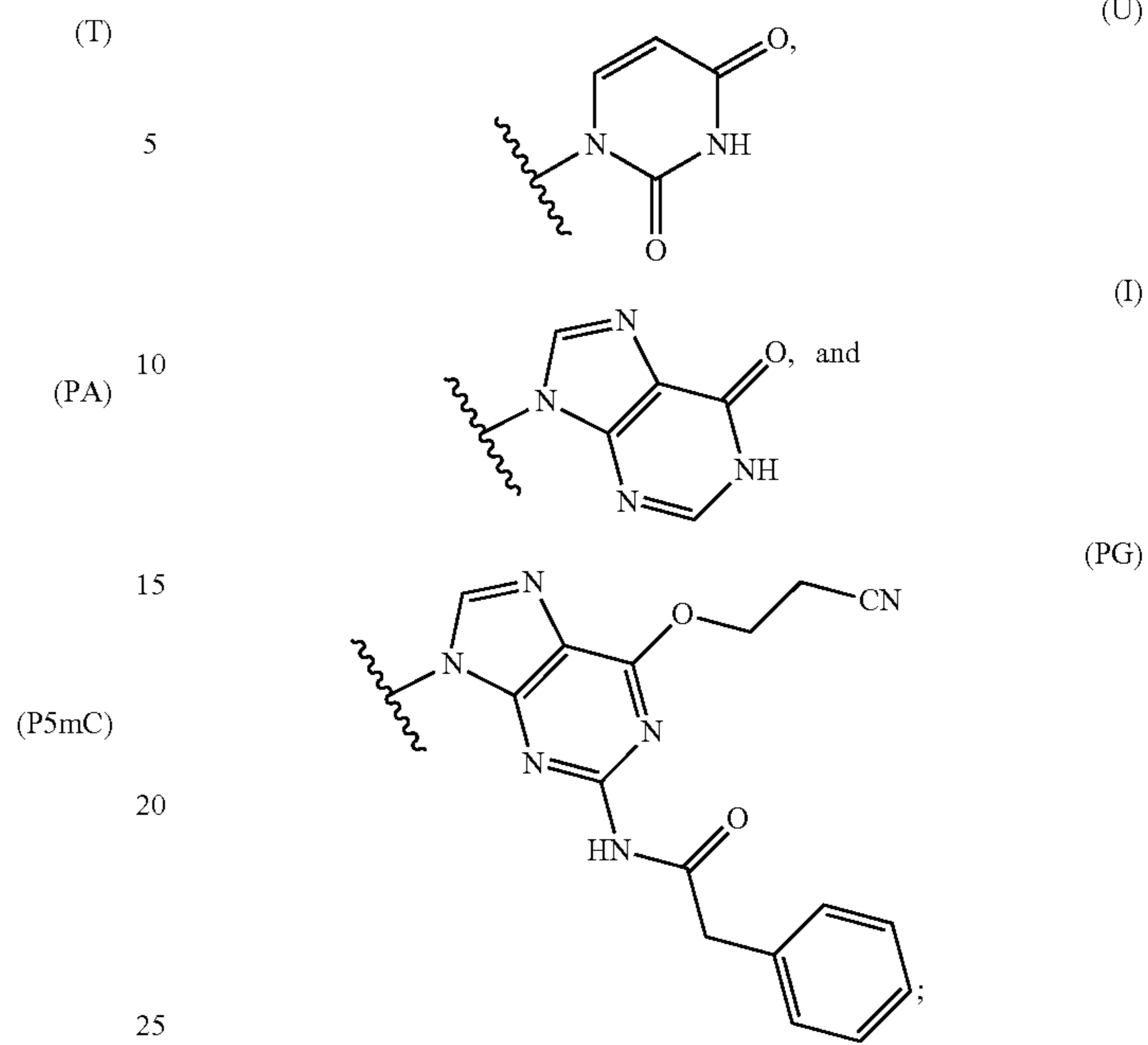
61

-continued

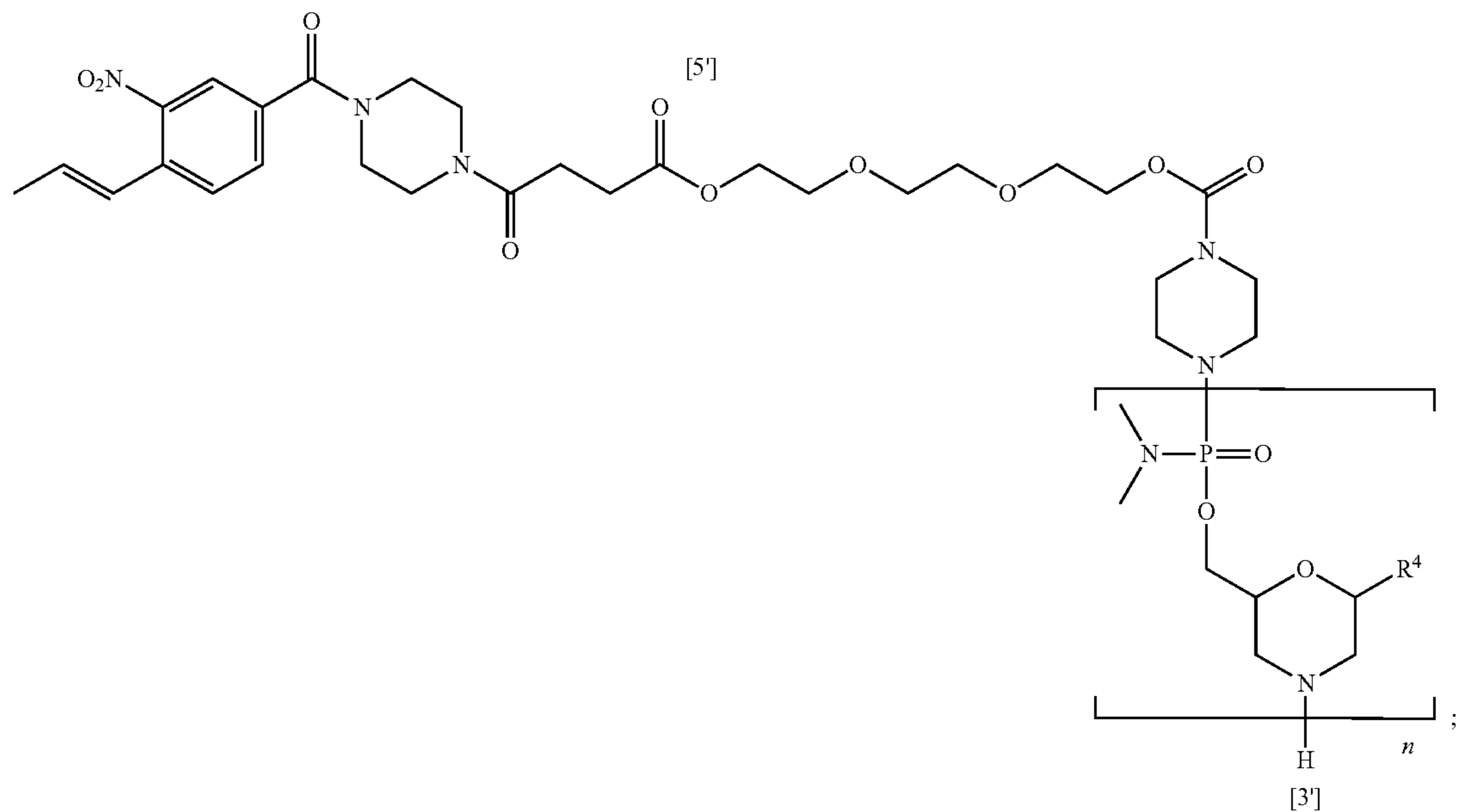


62

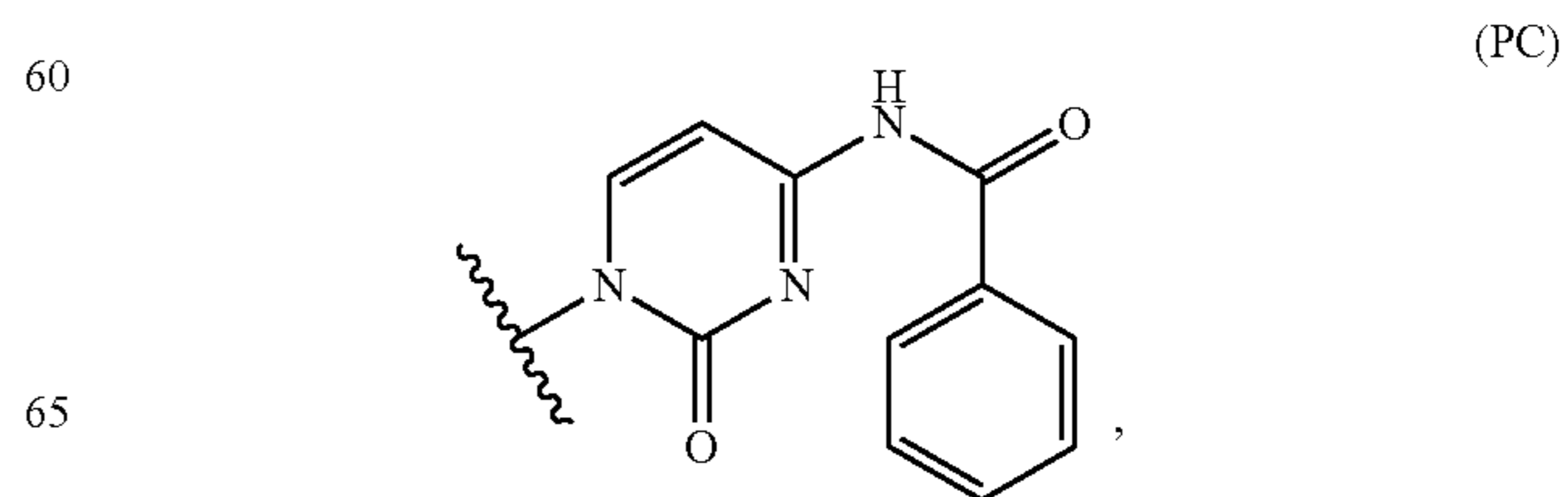
-continued



(g) contacting the compound of Formula (A10) with a cleaving agent to form a compound of Formula (A11):

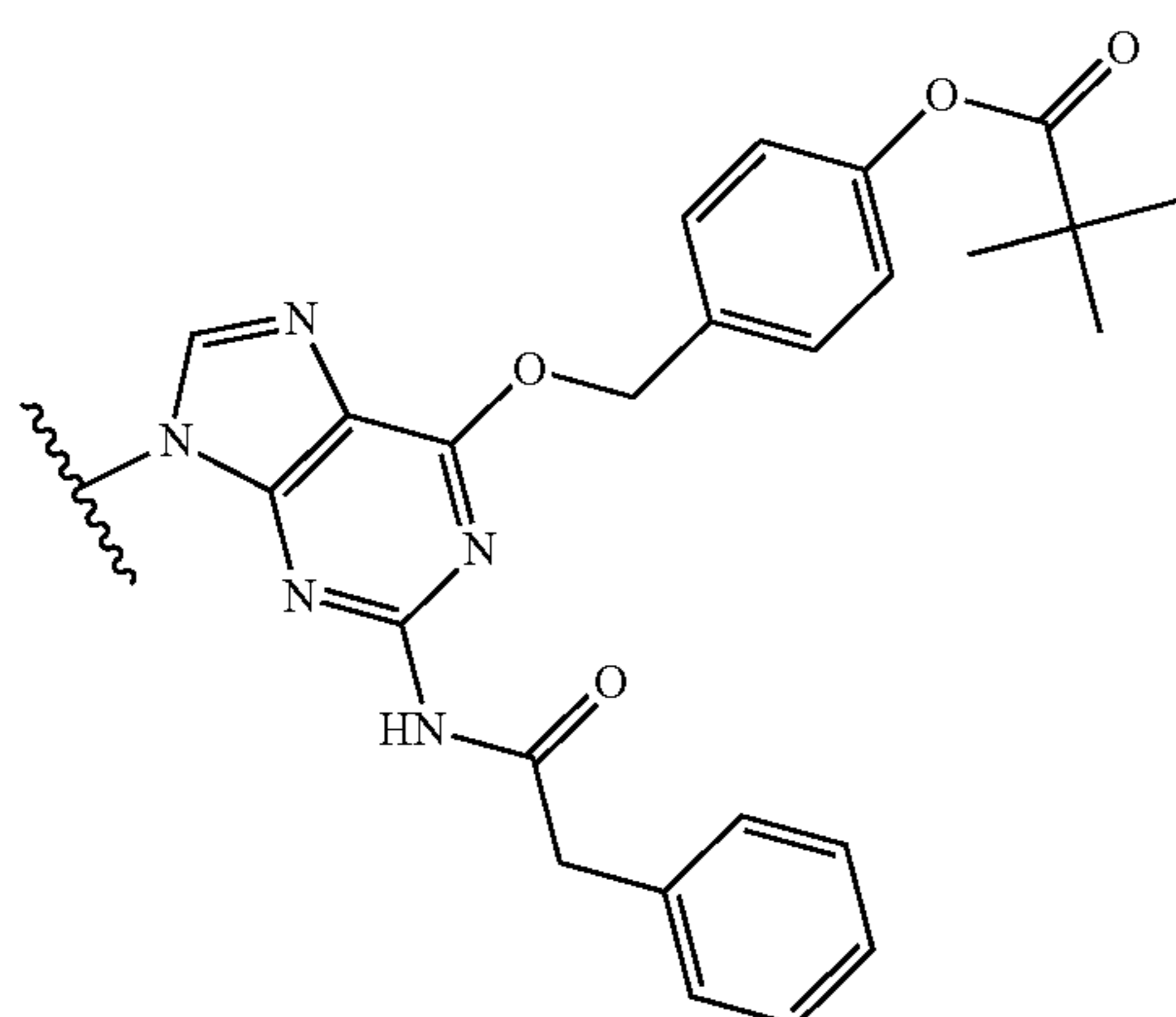


wherein n is an integer from 10 to 40, and R⁴ is, independently for each occurrence, selected from the group consisting of:

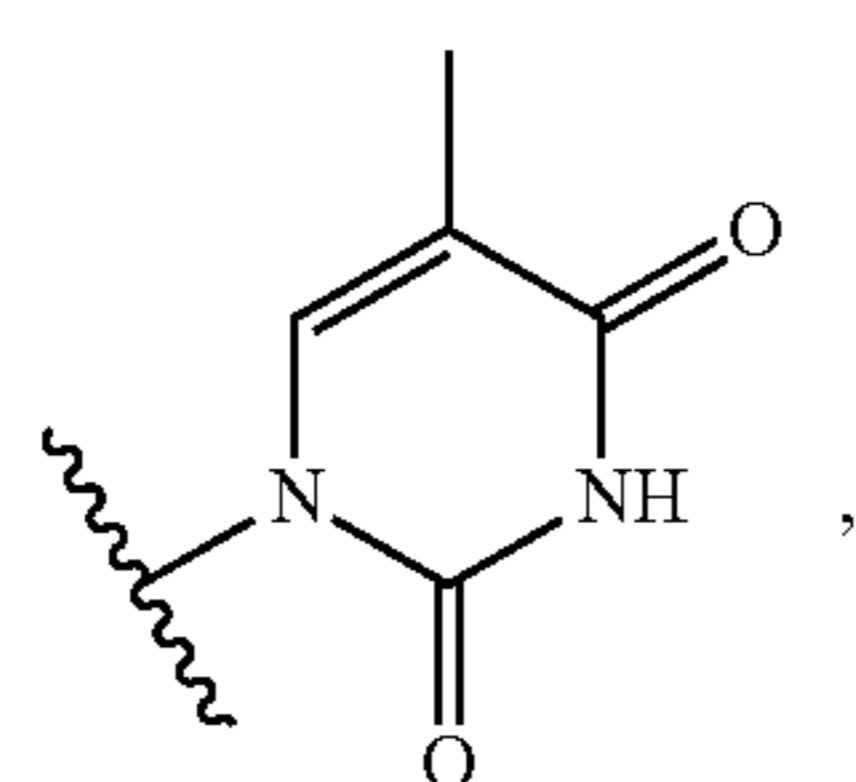


63

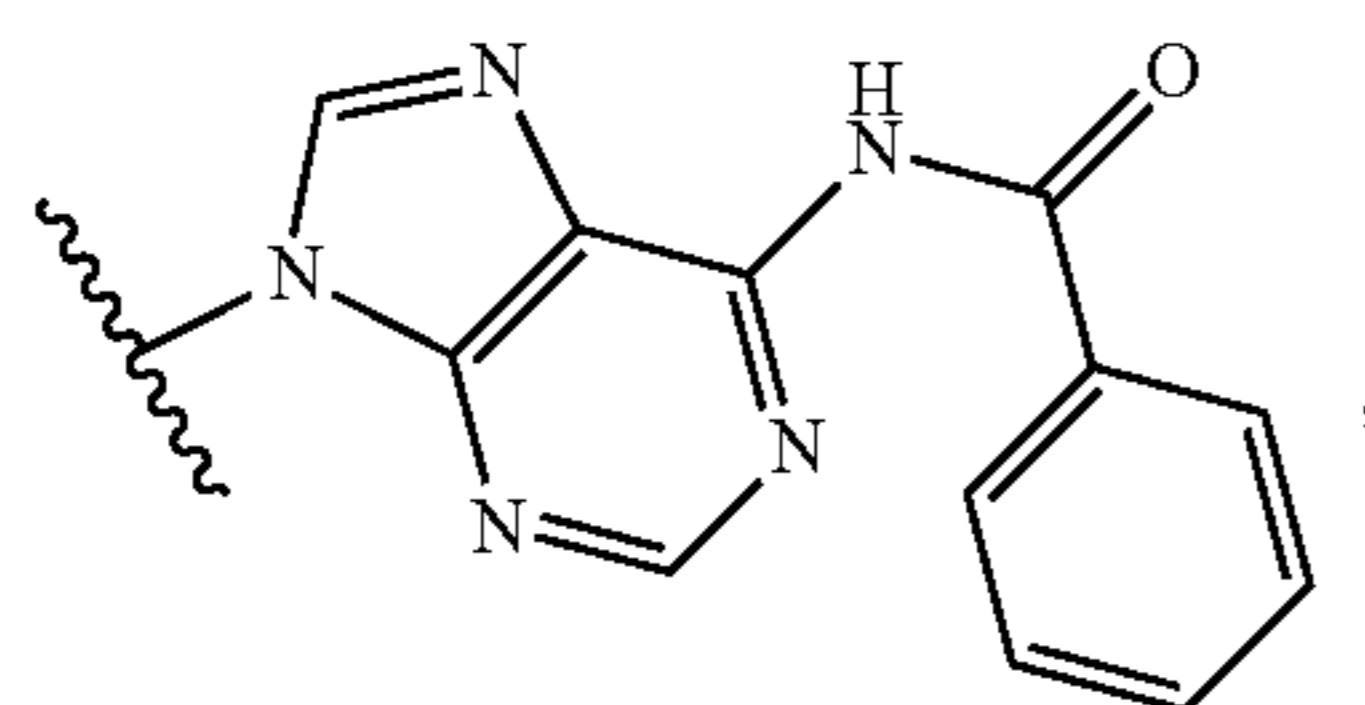
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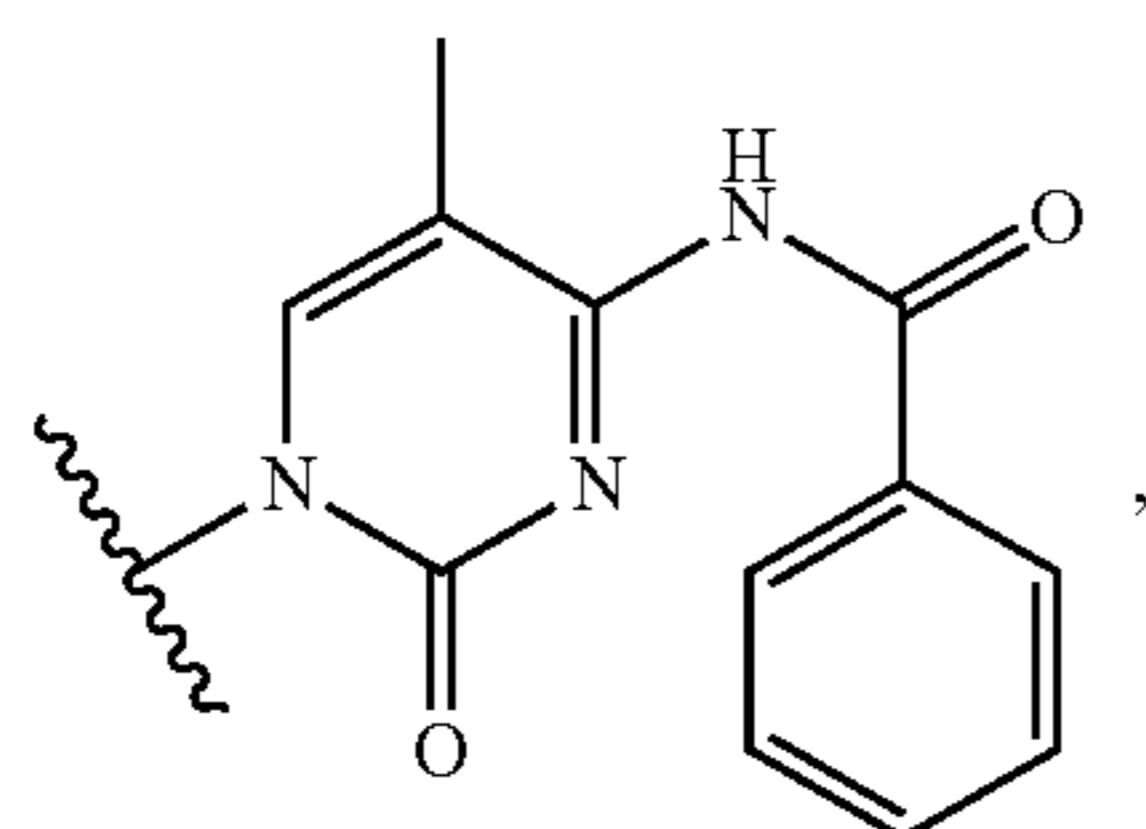
(DPG)



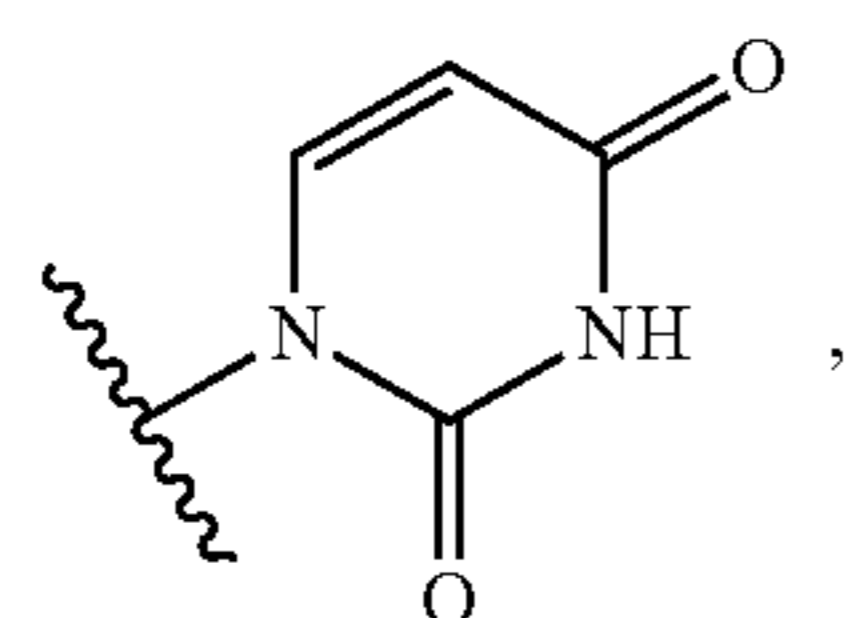
(T)



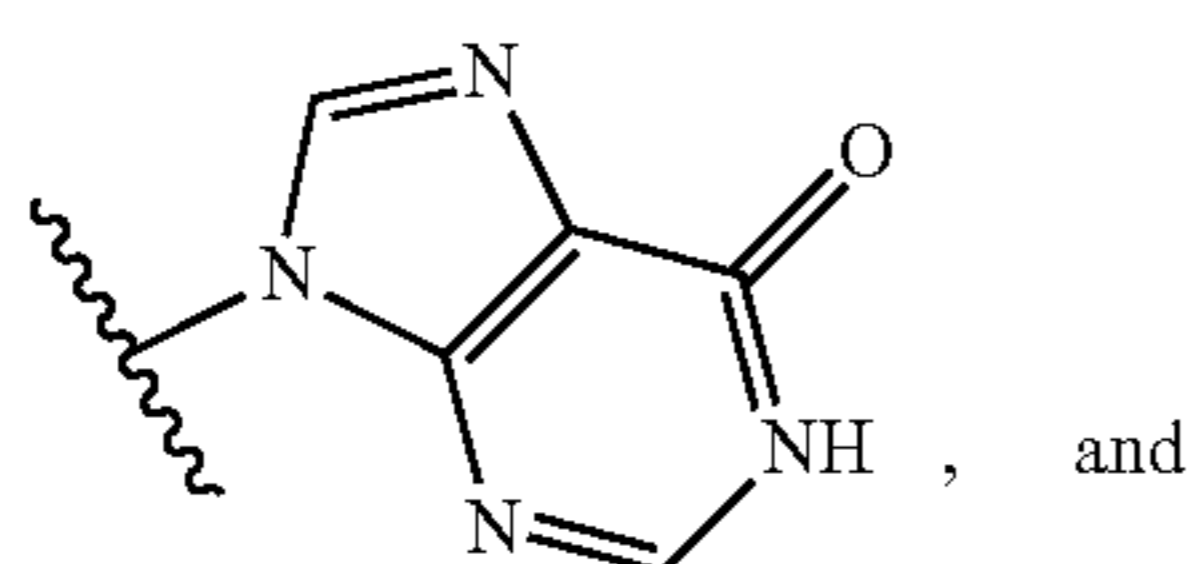
(PA)



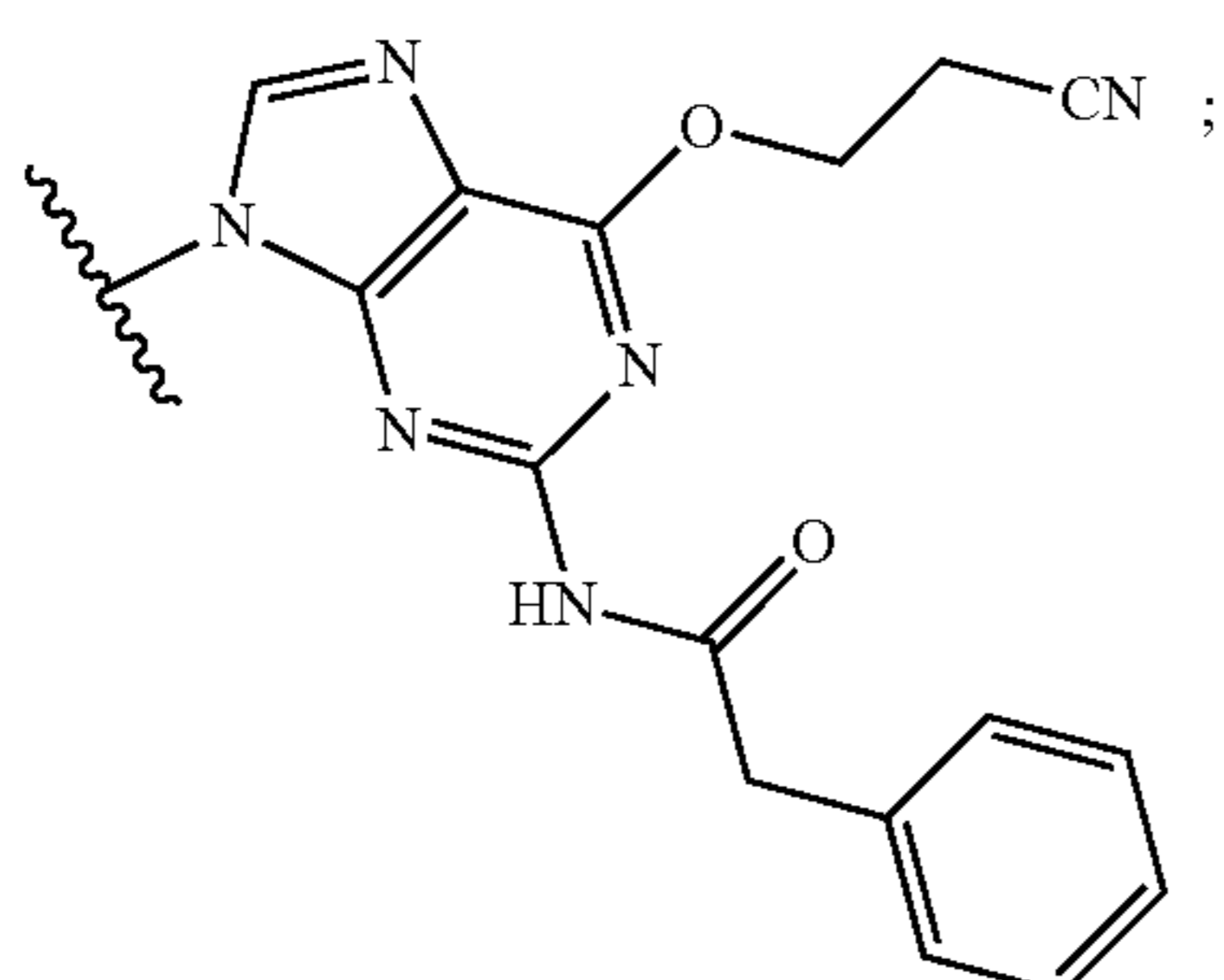
(P5mC)



(U)



(I)



(PG)

64

and

(h) contacting the compound of Formula (A11) with a deprotecting agent to form the oligomeric compound of Formula (A).

5

In one embodiment, step (d) or step (e2) further comprises contacting the compound of Formula (IV) or the compound formed by the immediately prior step, respectively, with a capping agent.

In another embodiment, each step is performed in the presence of at least one solvent.

In yet another embodiment, the deblocking agent used in each step is a solution comprising a halogenated acid.

In still another embodiment, the deblocking agent used in each step is cyanoacetic acid.

In another embodiment, the halogenated acid is selected from the group consisting of chloroacetic acid, dichloroacetic acid, trichloroacetic acid, fluoroacetic acid, difluoroacetic acid, and trifluoroacetic acid.

In another embodiment, the halogenated acid is trifluoroacetic acid.

In yet another embodiment, at least one of steps (a), (c), (e1), and (f) further comprises the step of contacting the deblocked compound of each step with a neutralization agent.

In still another embodiment, each of steps (a), (c), (e1), and (f) further comprises the step of contacting the deblocked compound of each step with a neutralization agent.

In another embodiment, the neutralization agent is in a solution comprising dichloromethane and isopropyl alcohol.

In yet another embodiment, the neutralization agent is a monoalkyl, dialkyl, or trialkyl amine.

In still another embodiment, the neutralization agent is N,N-diisopropylethylamine.

In another embodiment, the deblocking agent used in each step is a solution comprising 4-cyanopyridine, dichloromethane, trifluoroacetic acid, trifluoroethanol, and water.

In yet another embodiment, the capping agent is in a solution comprising ethylmorpholine and methylpyrrolidone.

In still another embodiment, the capping agent is an acid anhydride.

In another embodiment, the acid anhydride is benzoic anhydride.

In another embodiment, the compounds of Formula (A4) and Formula (A8) are each, independently, in a solution comprising ethylmorpholine and dimethylimidazolidinone.

In another embodiment, the cleavage agent comprises dithiothreitol and 1,8-diazabicyclo[5.4.0]undec-7-ene.

In still another embodiment, the cleavage agent is in a solution comprising N-methyl-2-pyrrolidone.

In yet another embodiment, the deprotecting agent comprises NH_3 .

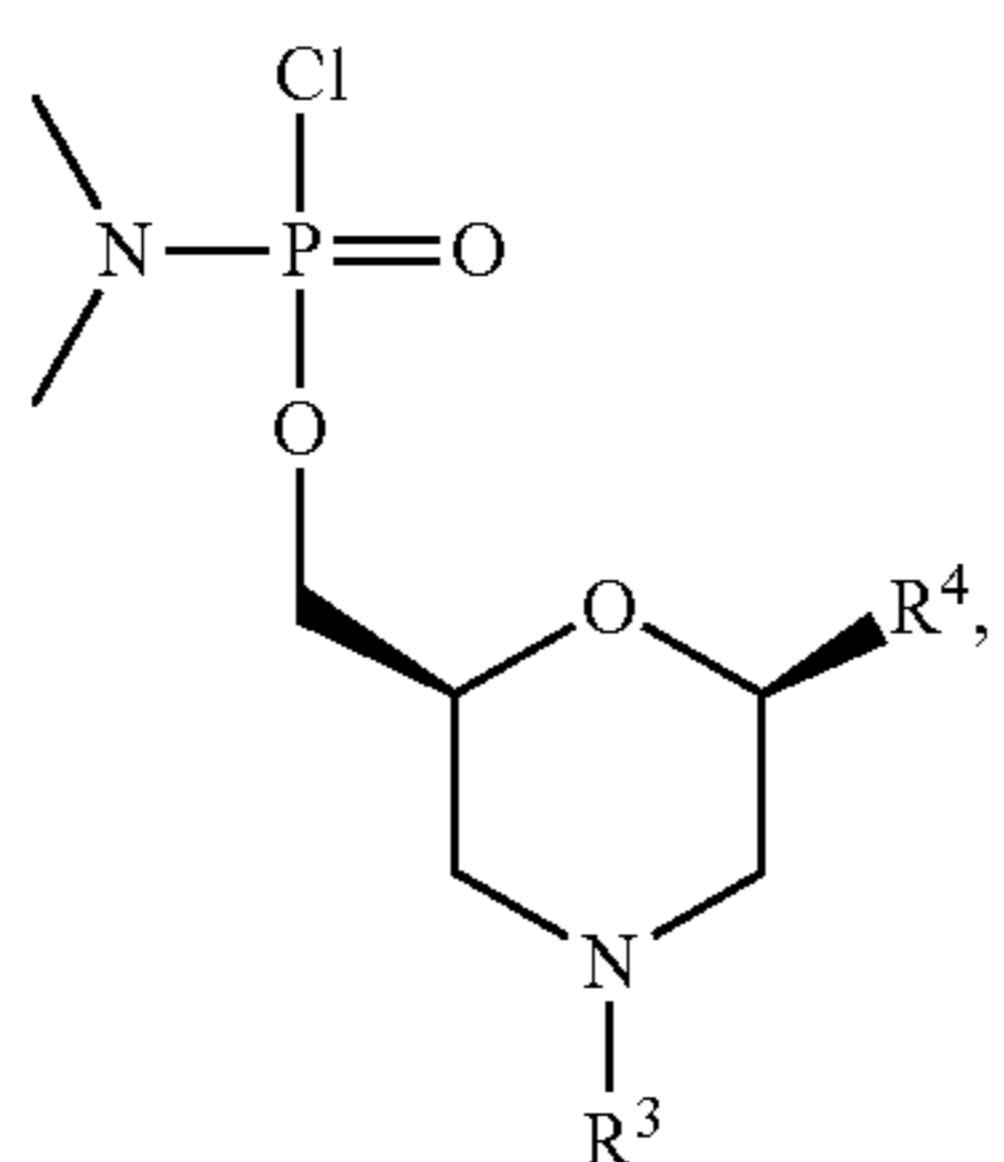
In still another embodiment, the deprotecting agent is in an aqueous solution.

In yet another embodiment, the support-medium comprises polystyrene with 1% crosslinked divinylbenzene.

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In another embodiment, the compound of Formula (A4) is of Formula (A4a):



(A4a) 5

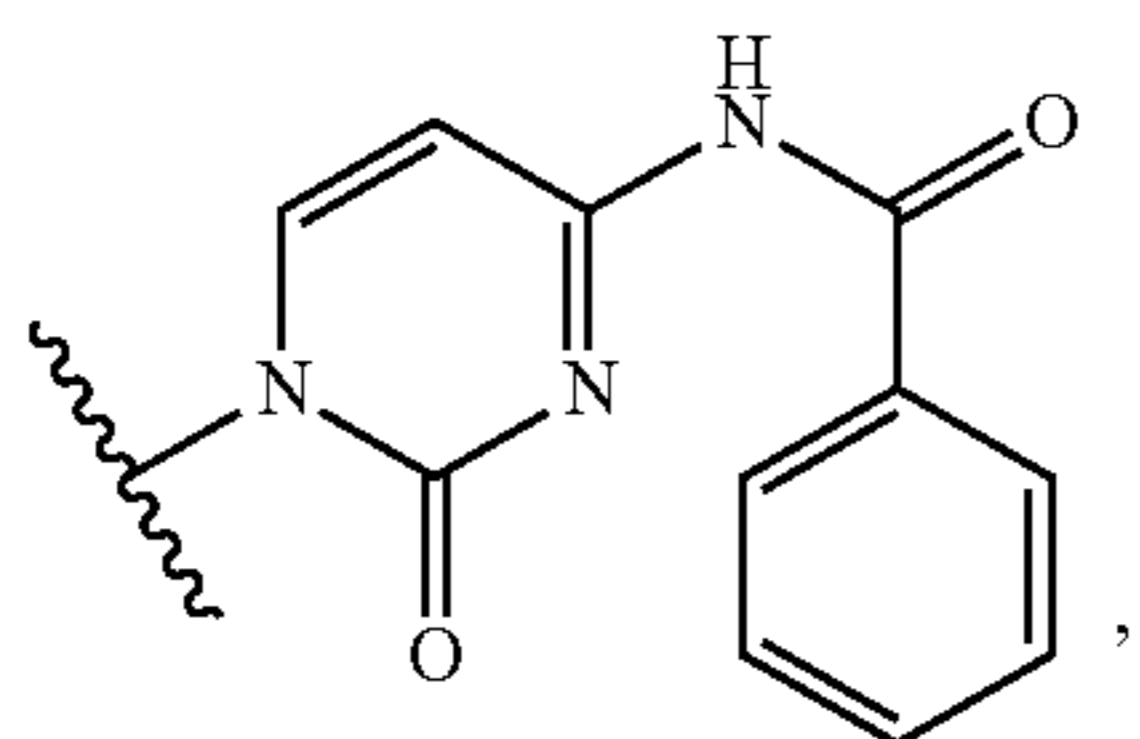
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wherein:

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

R⁴ is selected from:



(PC)

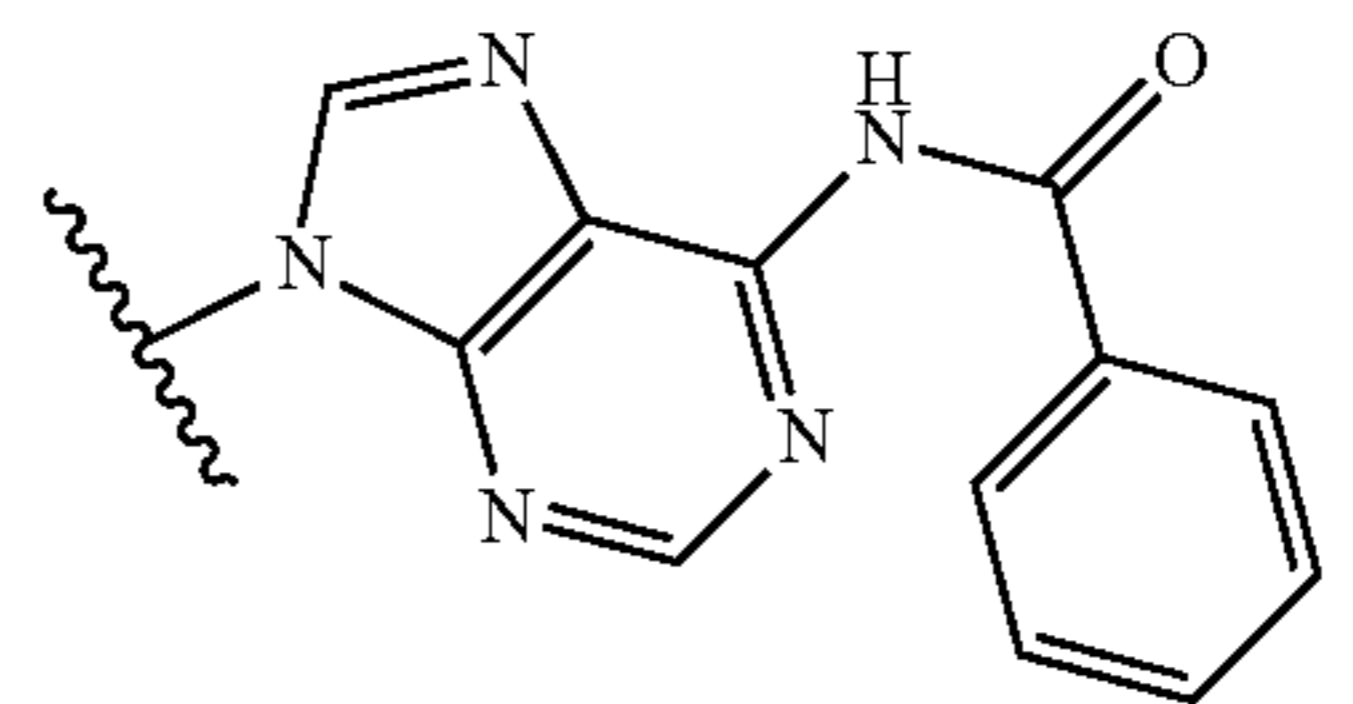
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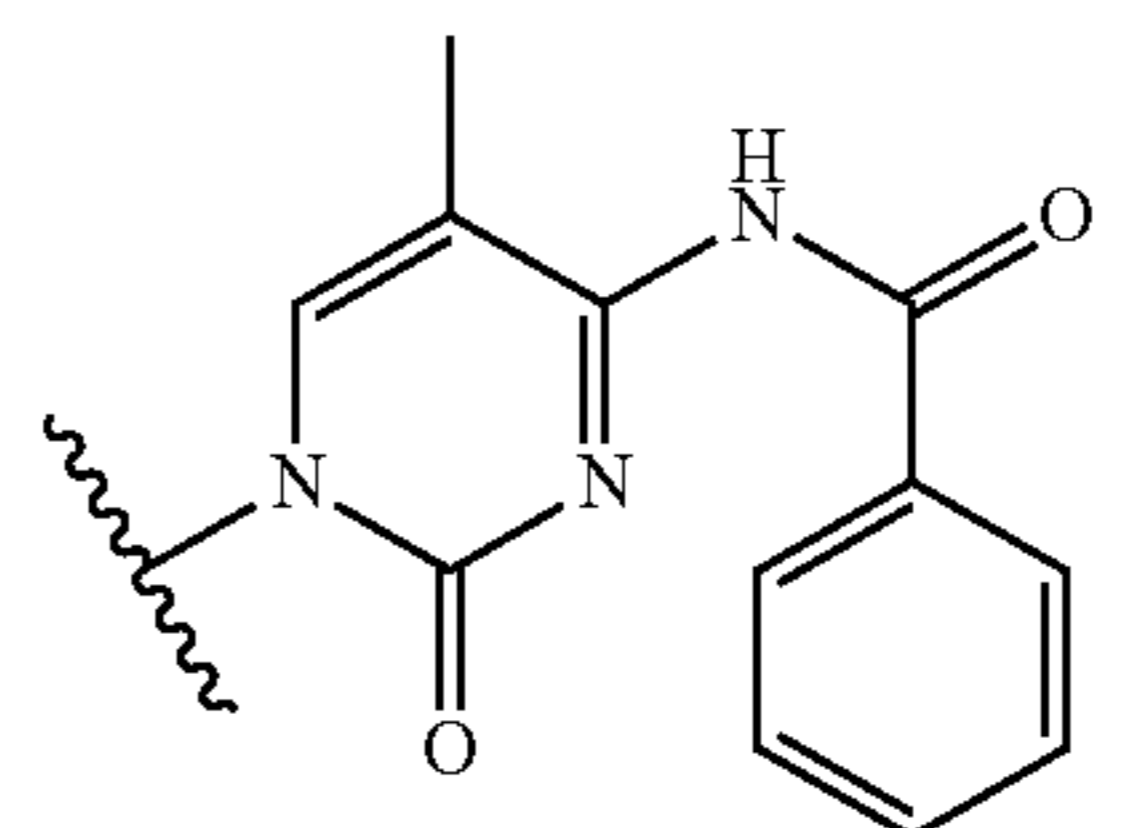
35

66

-continued



(PA)

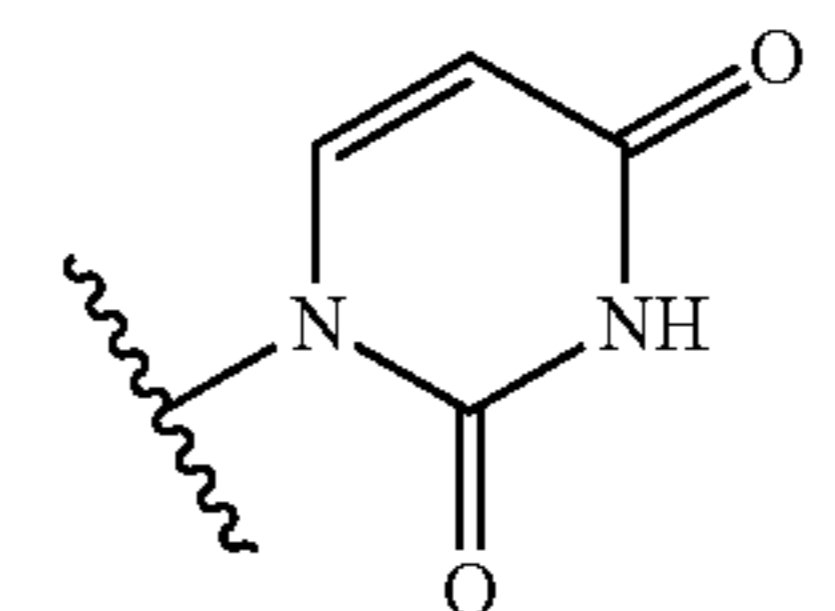


(P5mC)

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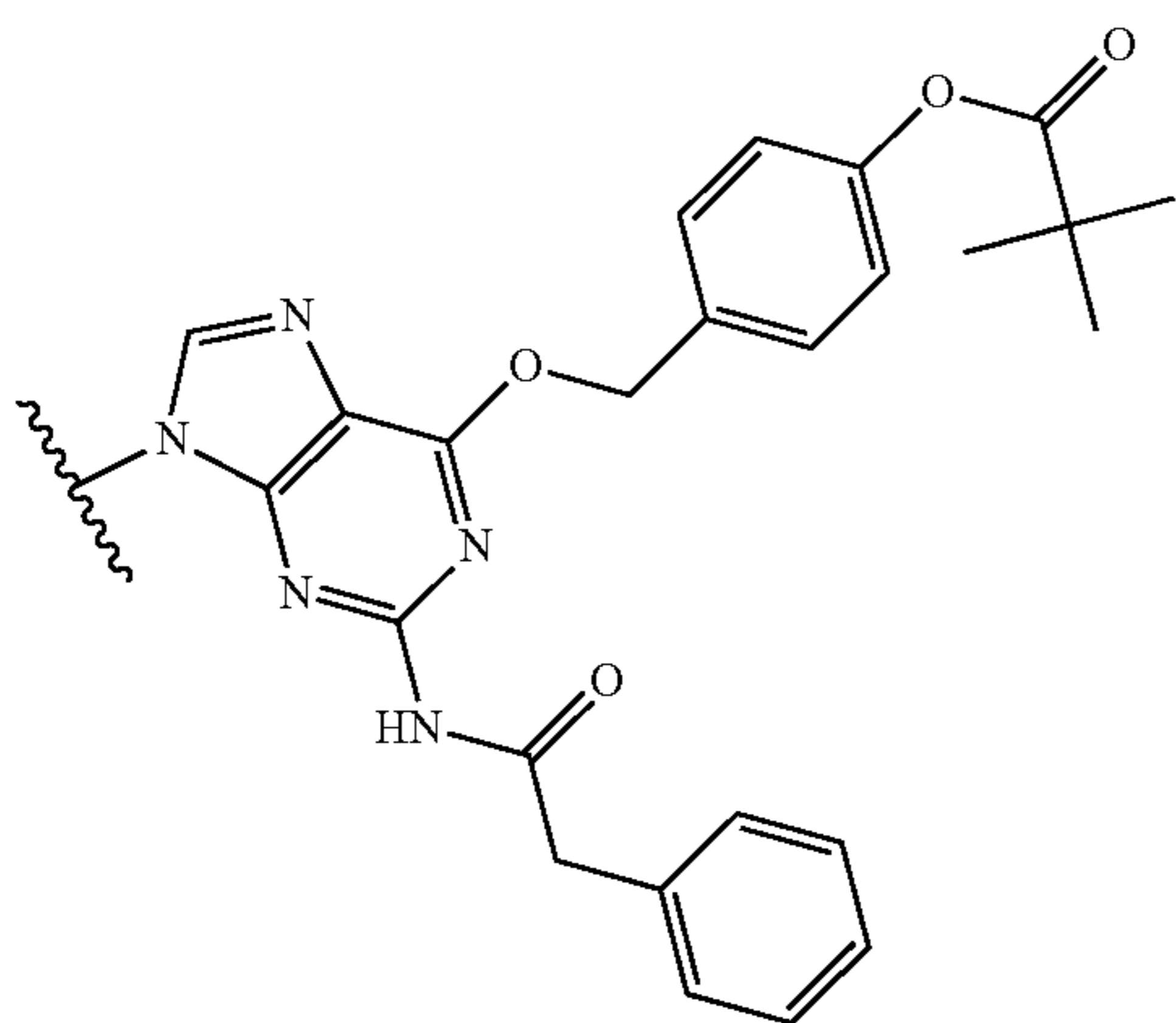
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(U)

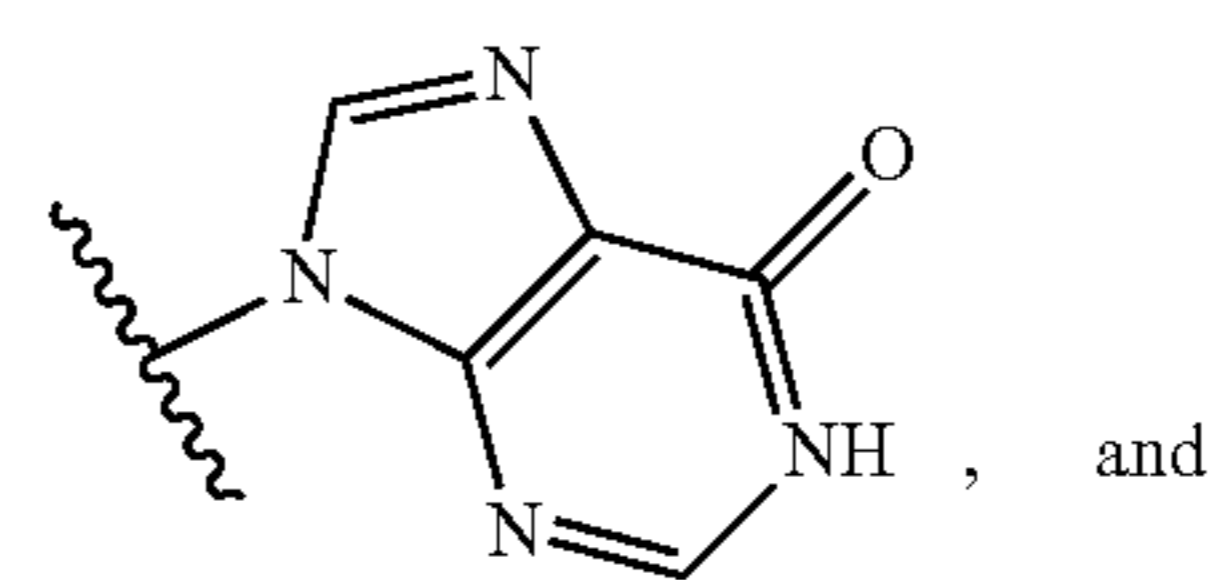
(DPG)



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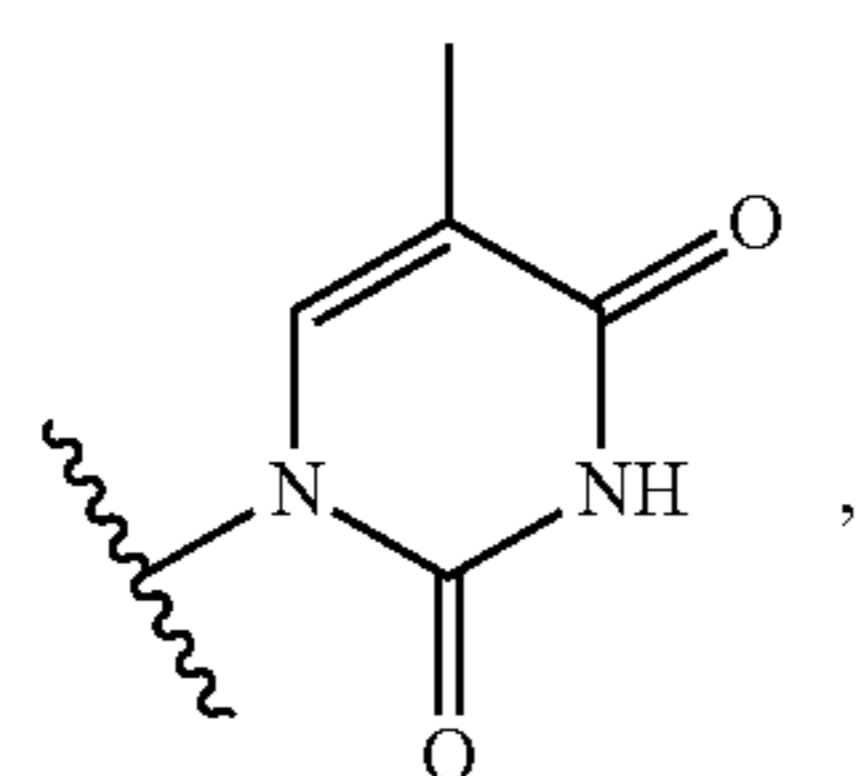
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(I)

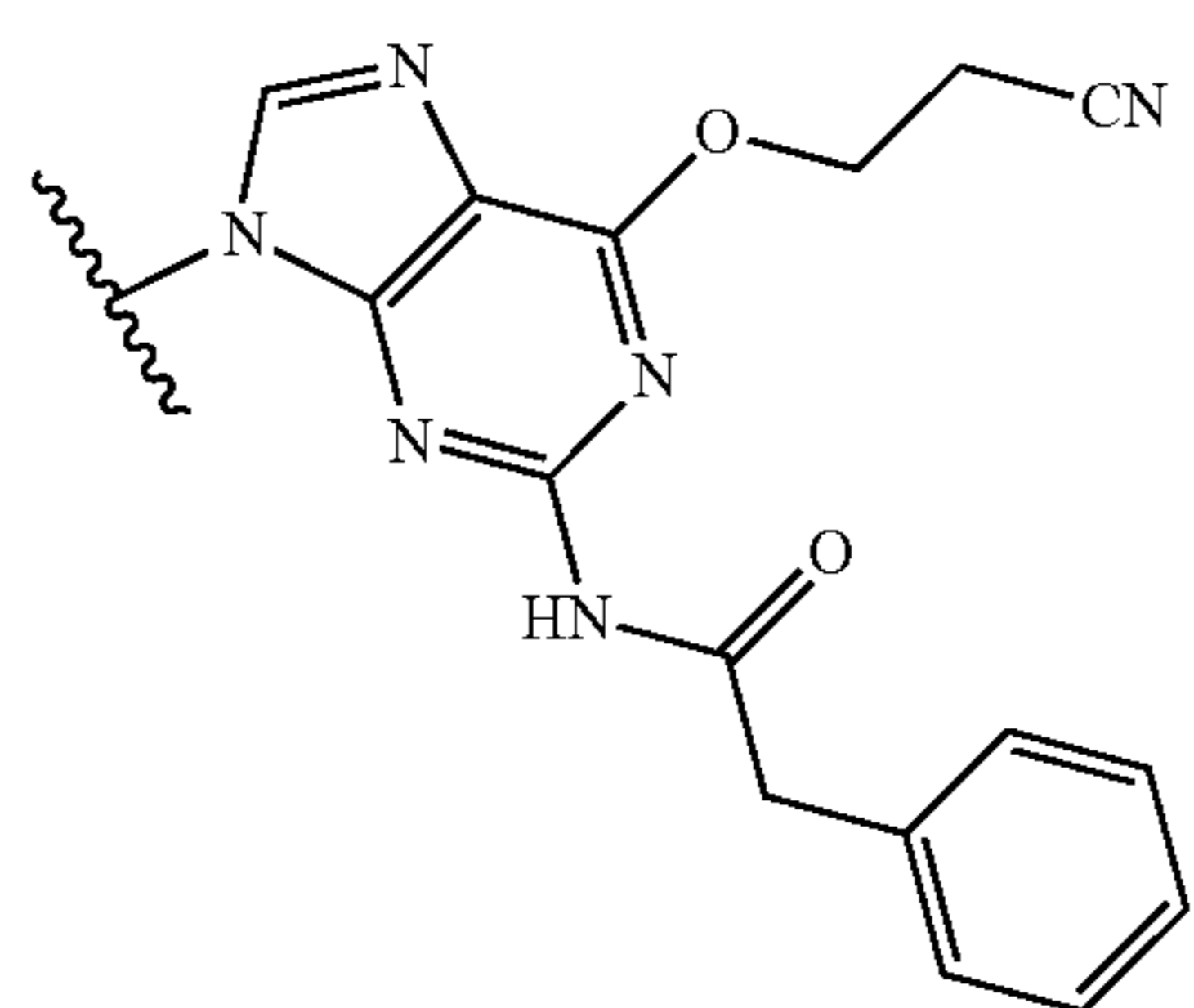
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(T)



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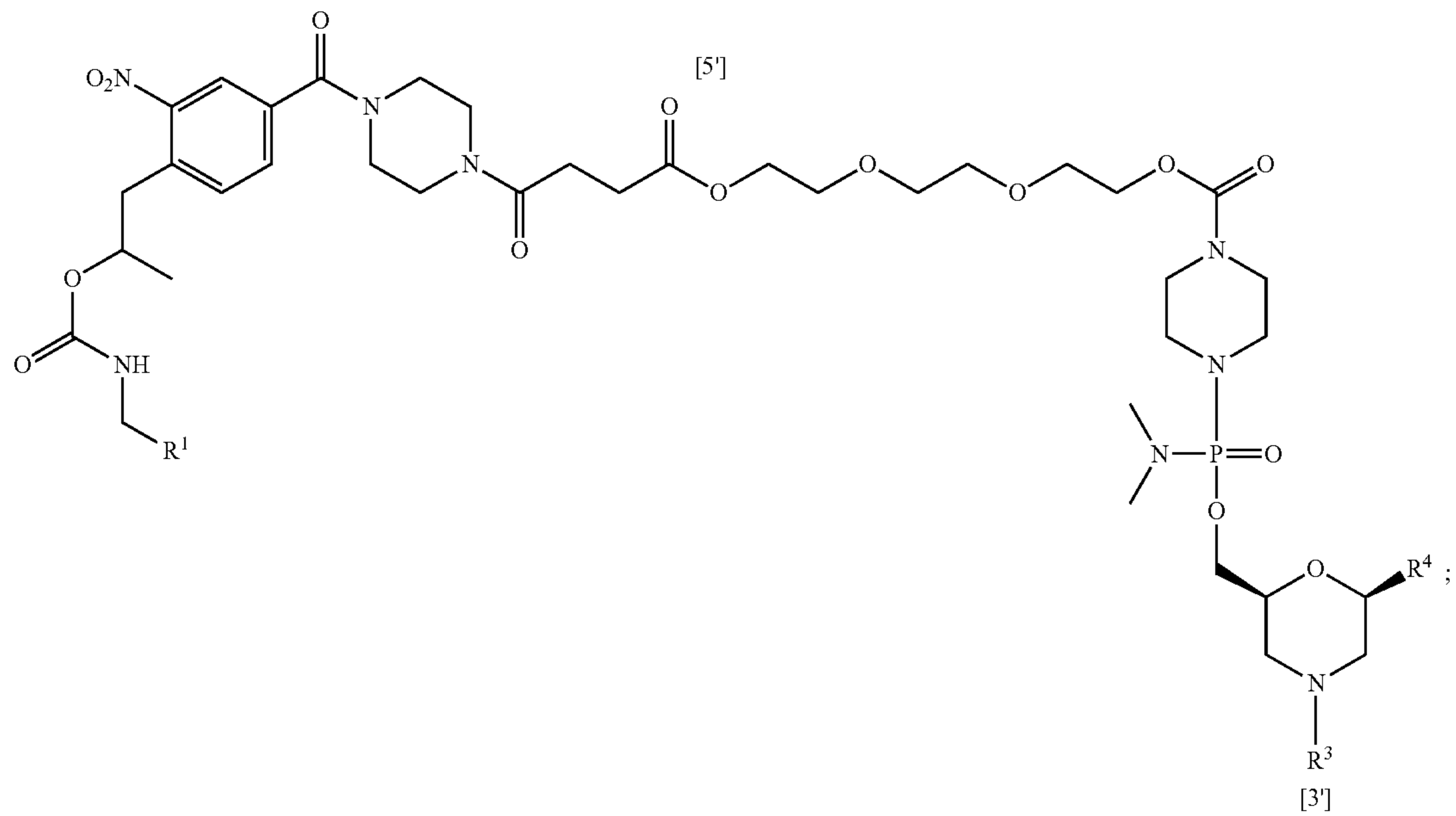


(PG)

67

In another embodiment, the compound of Formula (A5) is of Formula (A5a):

68

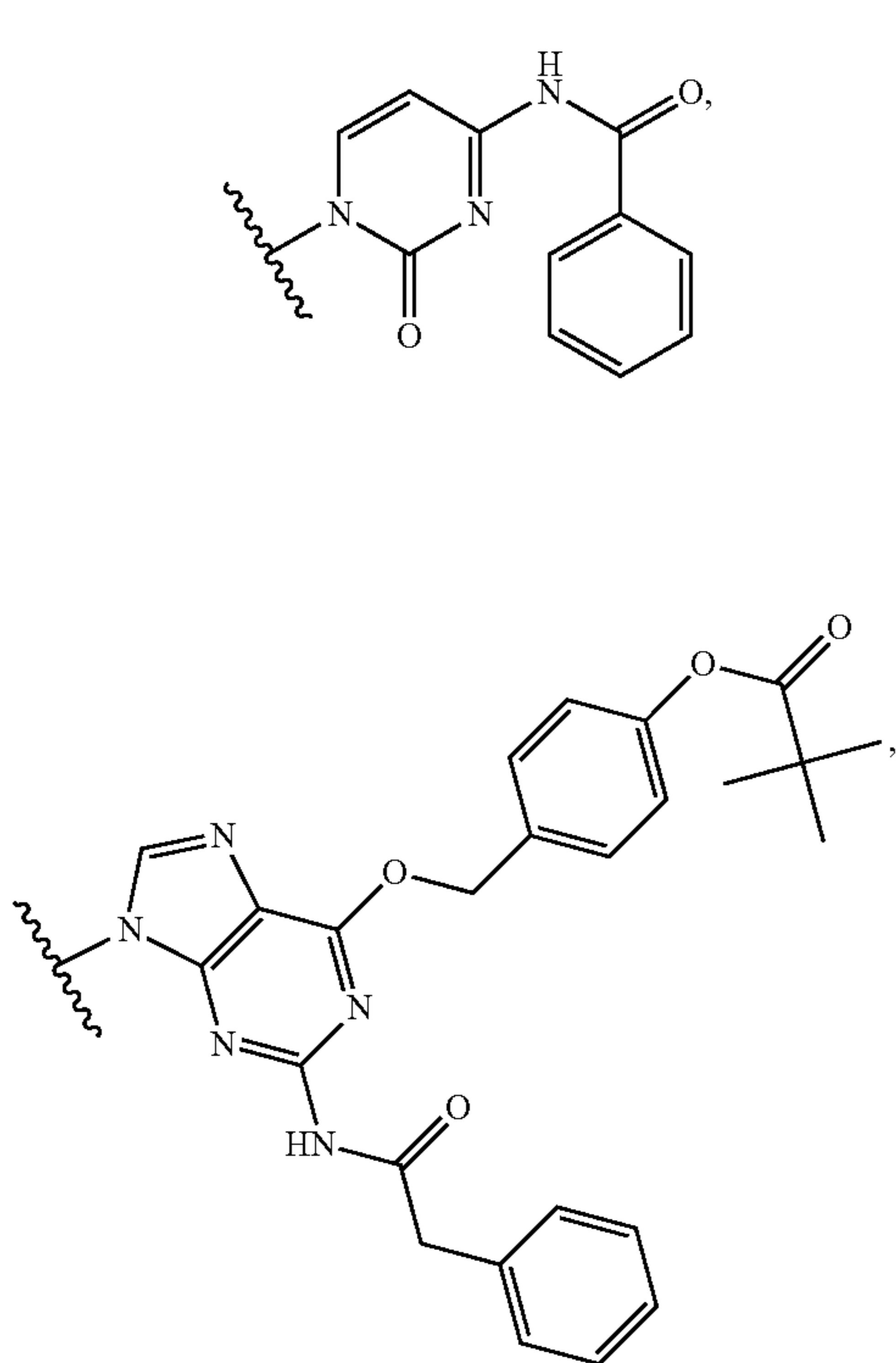


wherein:

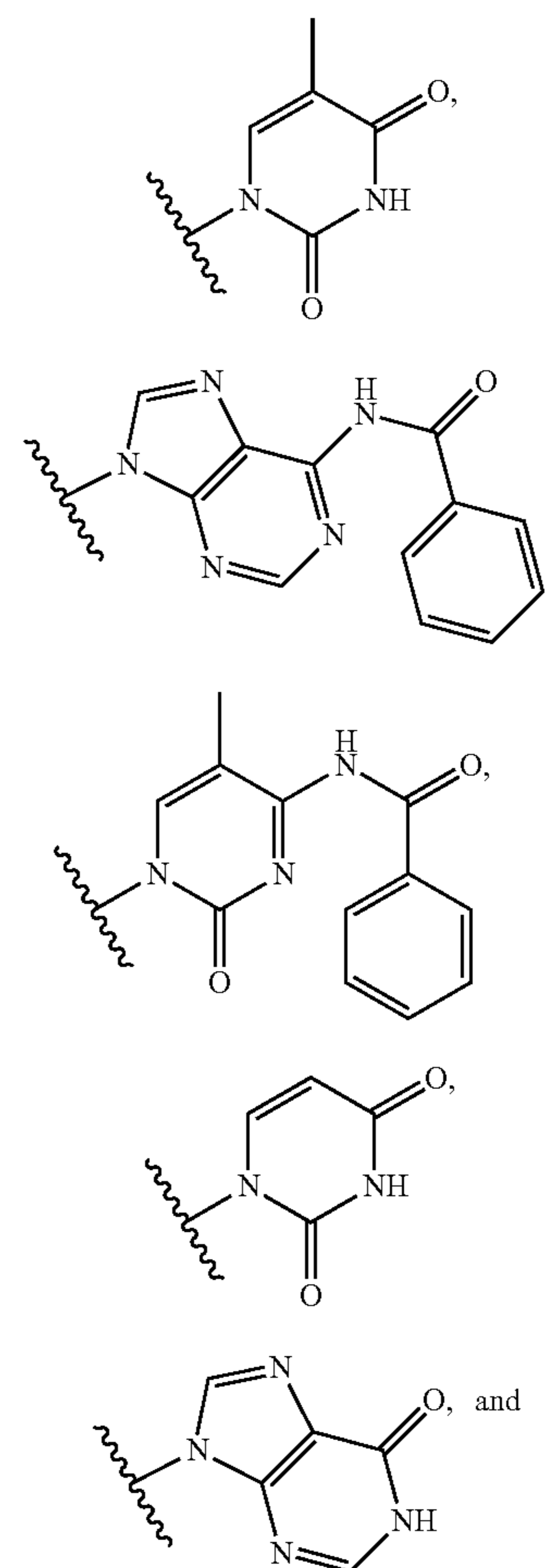
R¹ is a support-medium

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

R⁴ is selected from:

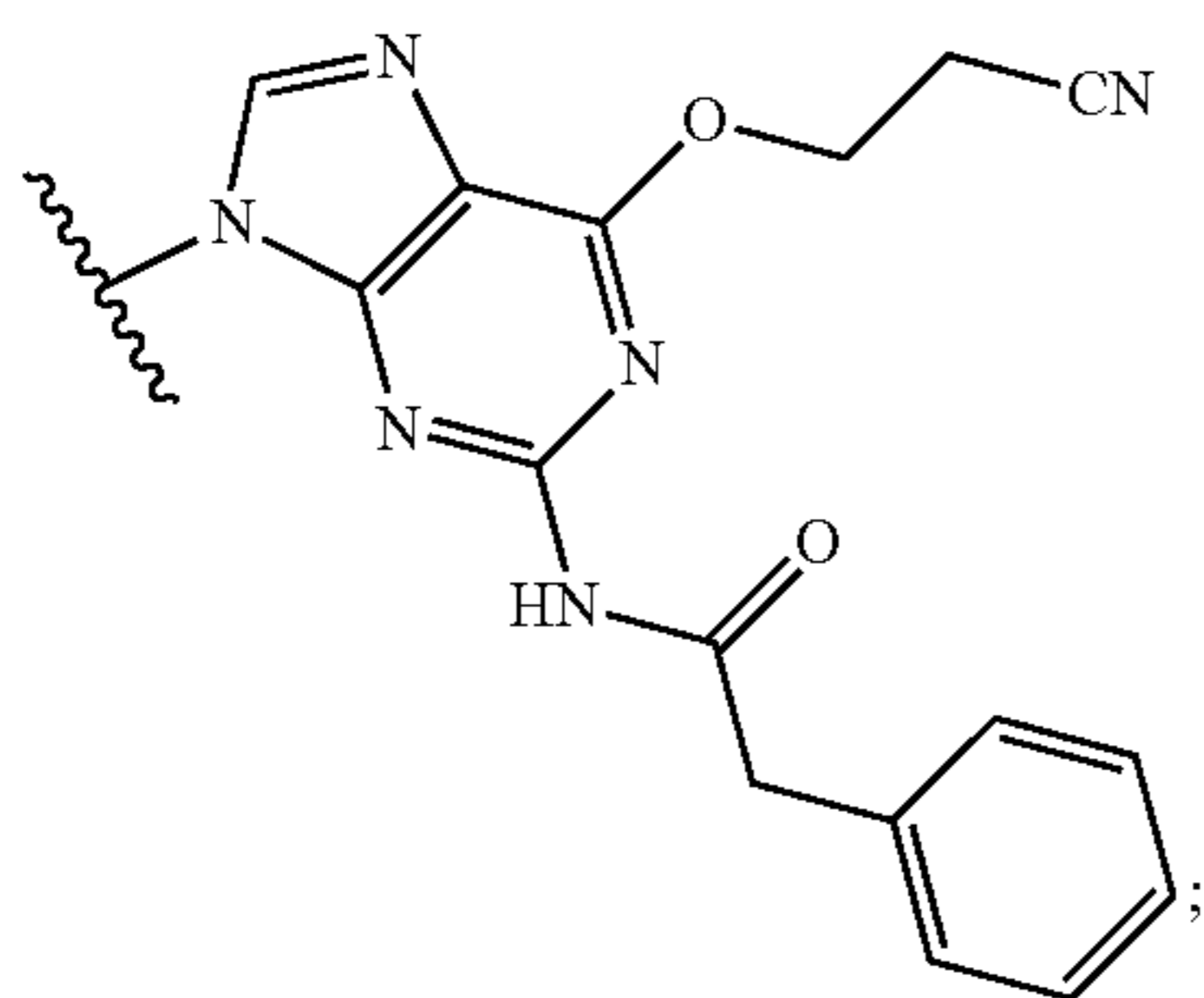


-continued



69

-continued



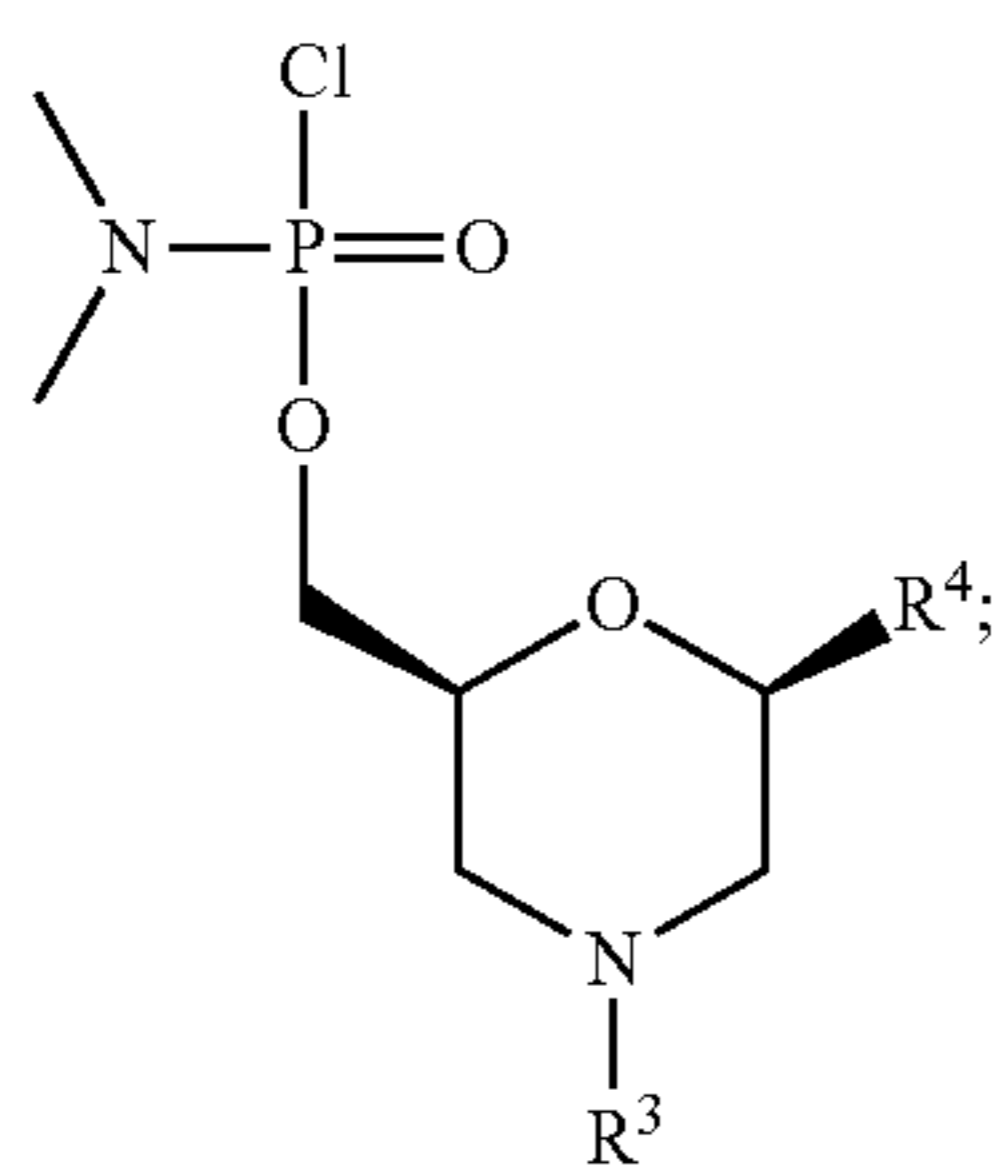
(PG)

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In yet another embodiment, the compound of Formula (A8) is of Formula (A8a):



(A8a)

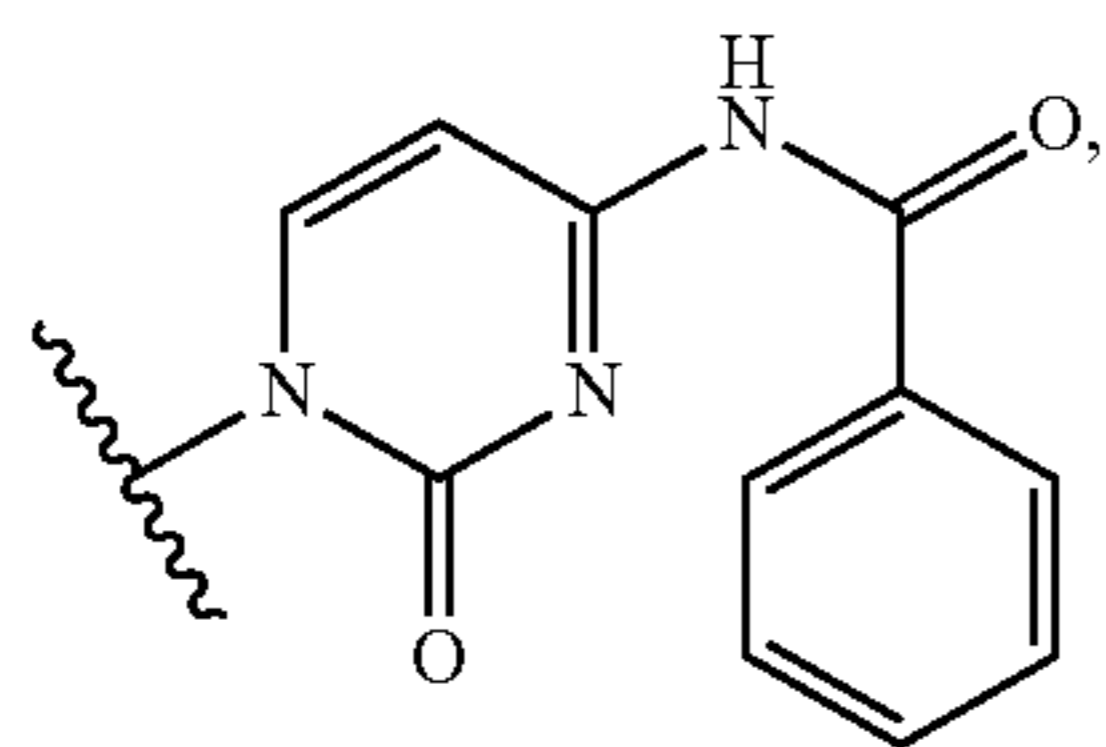
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wherein:

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

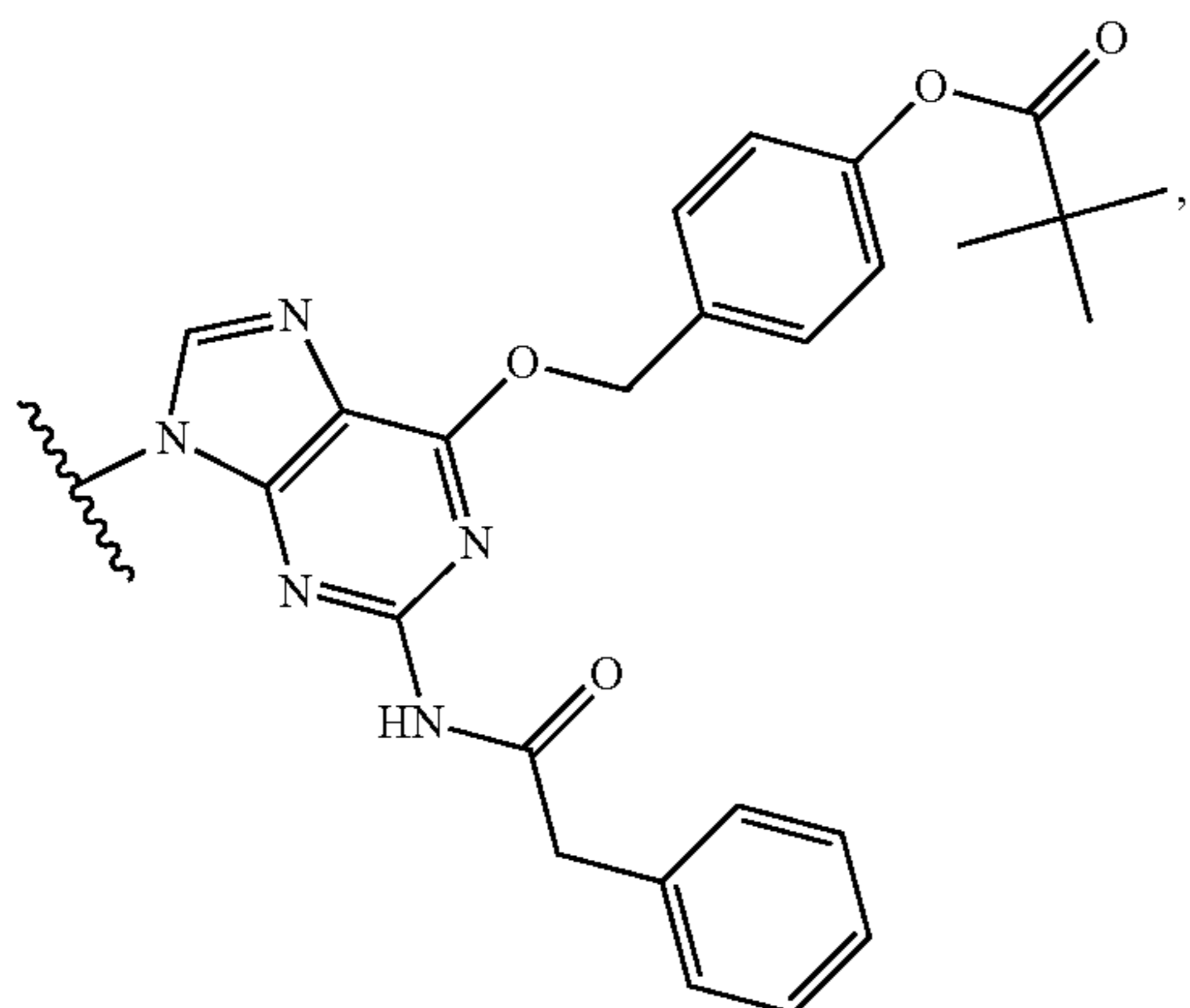
R⁴ is, independently at each occurrence of the compound of Formula (A8a), selected from the group consisting of:



(PC)

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(DPG)



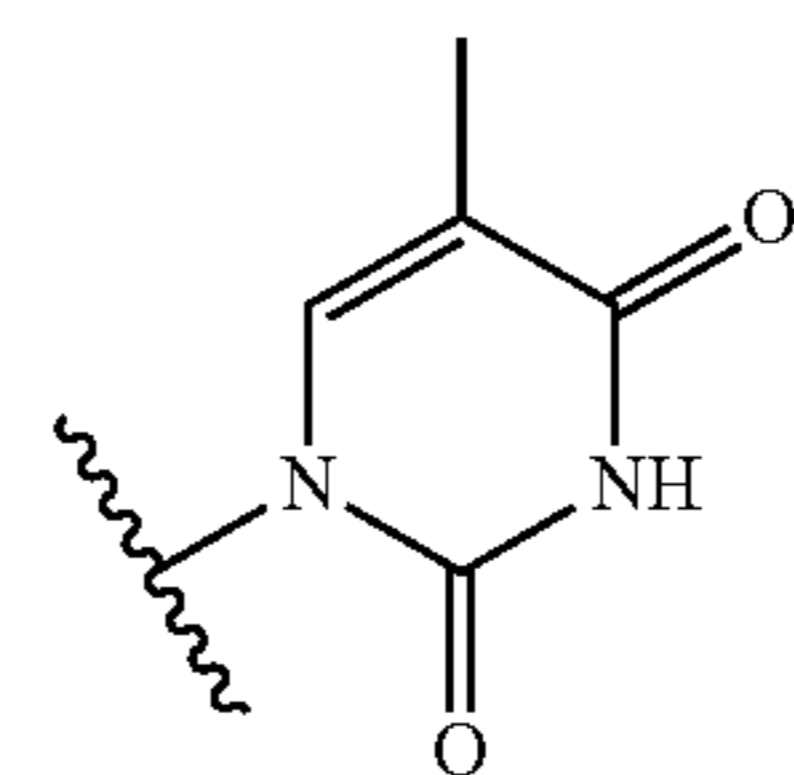
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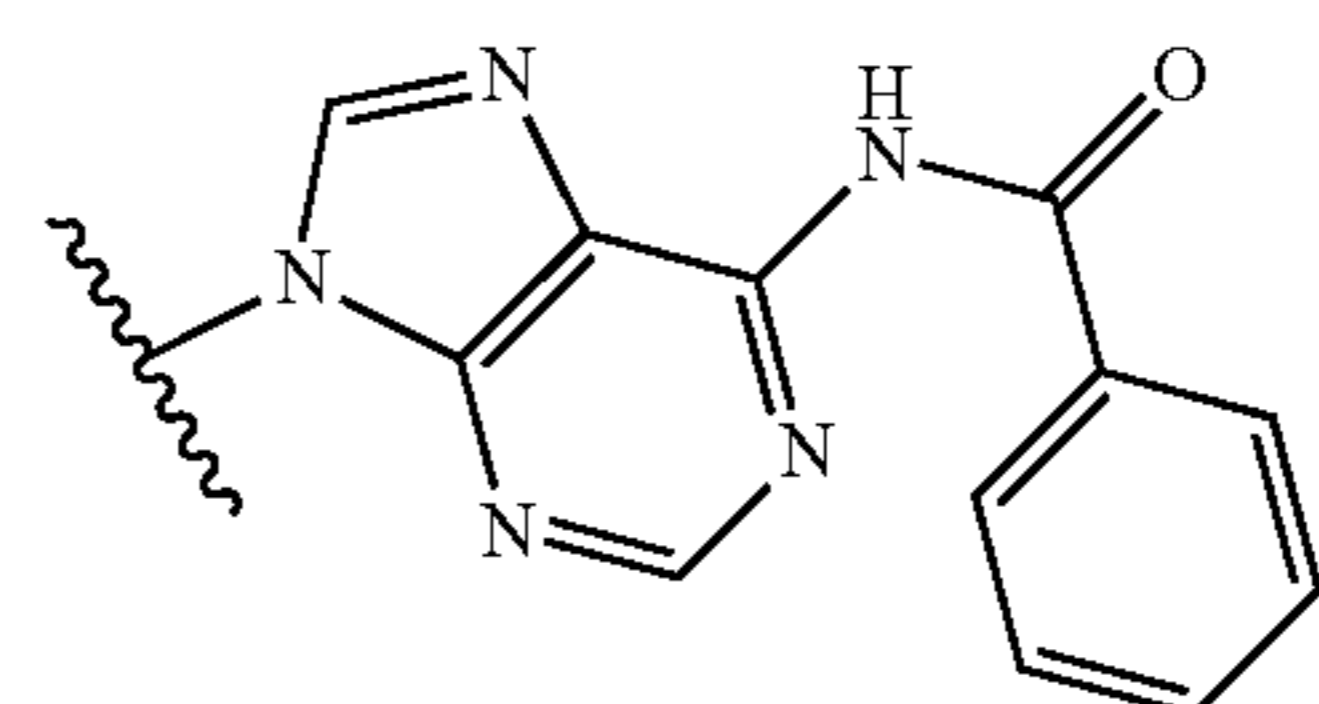
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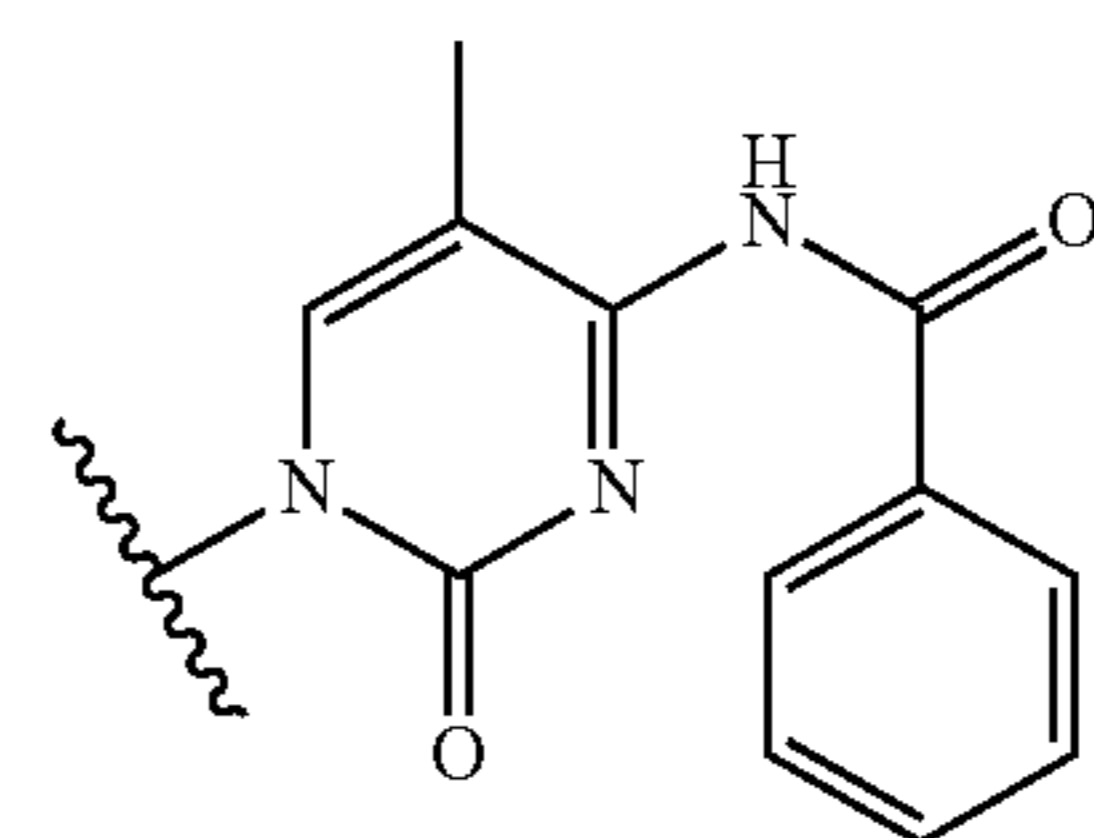
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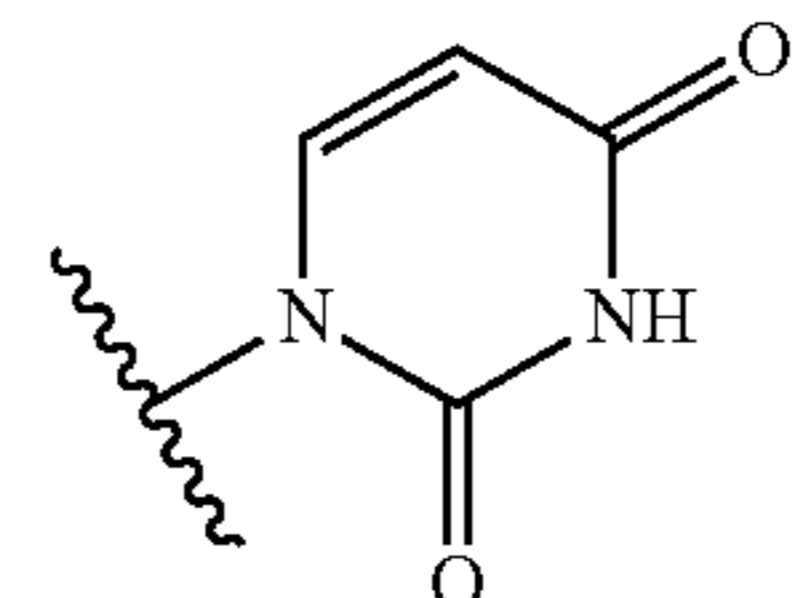
(T)



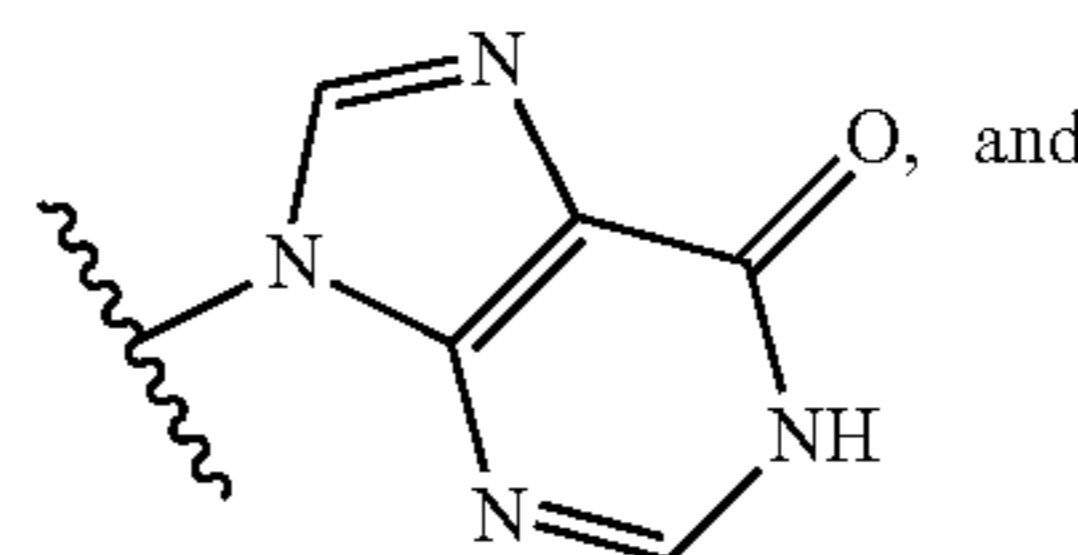
(PA)



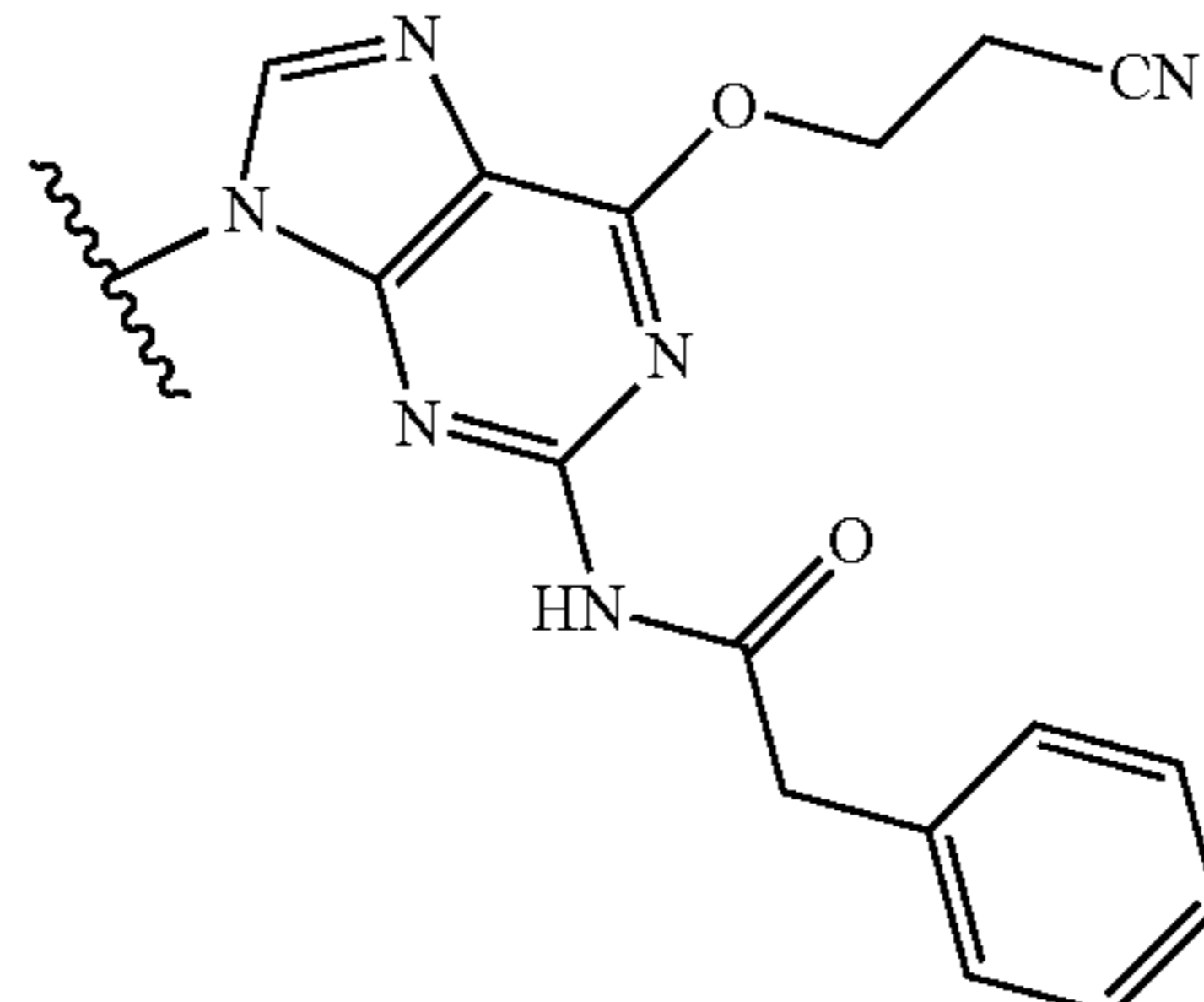
(P5mC)



(U)



(I)

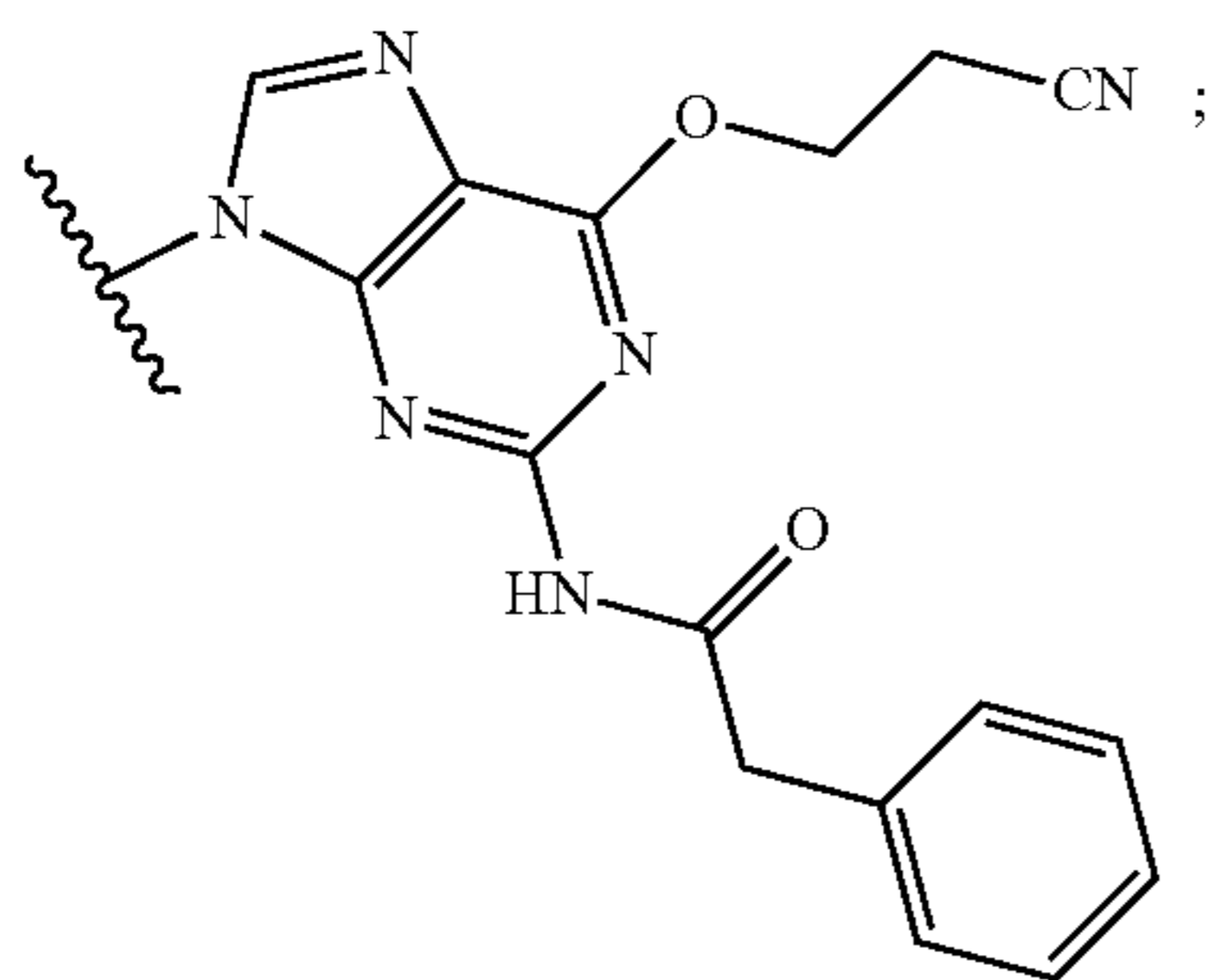


(PG)

In still another embodiment, the compound of formula (A9) is of Formula (A9a):

73

-continued

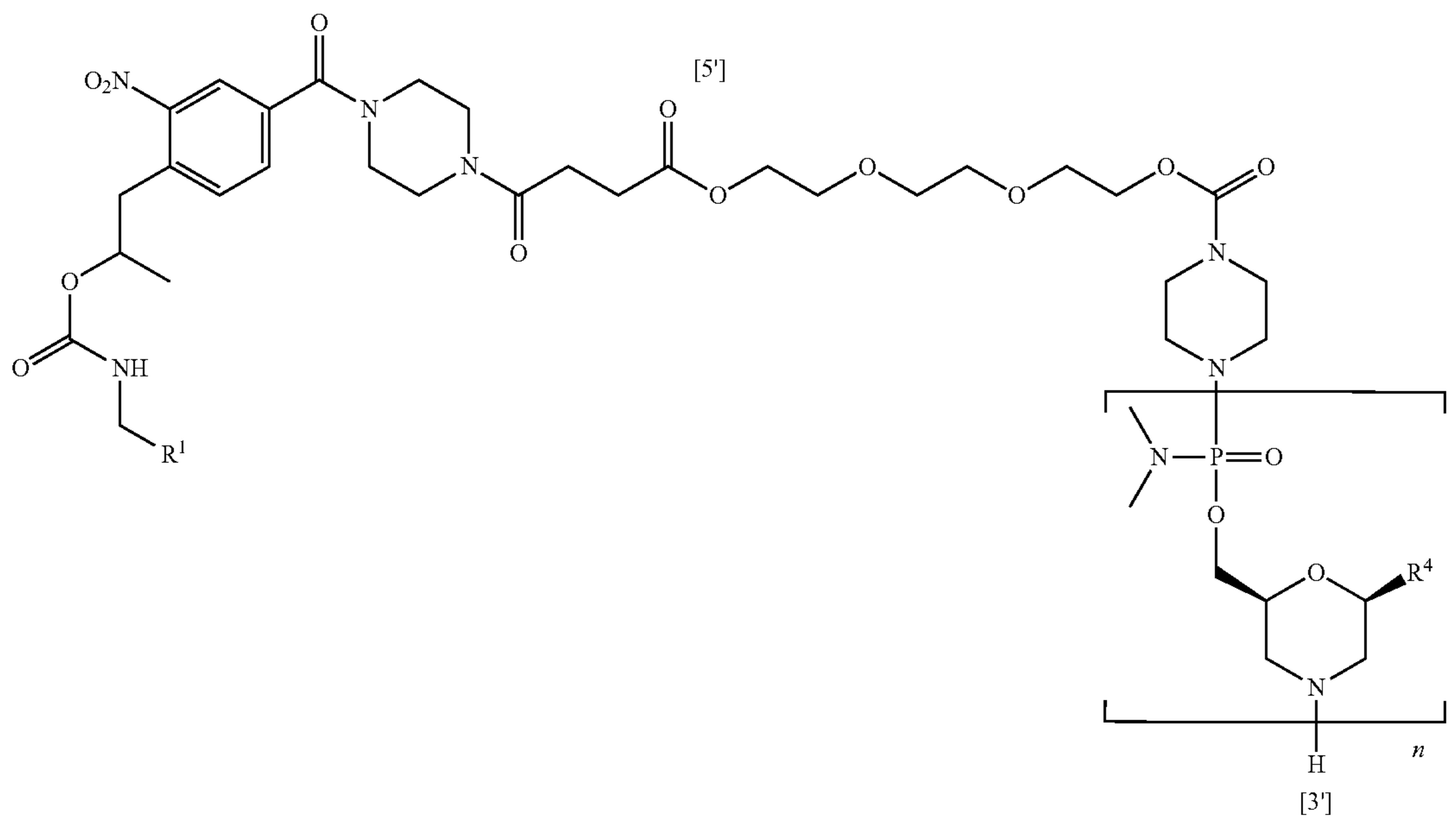


(PG)

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In another embodiment, the compound of Formula (A10) ¹⁵
is of Formula (A10a):



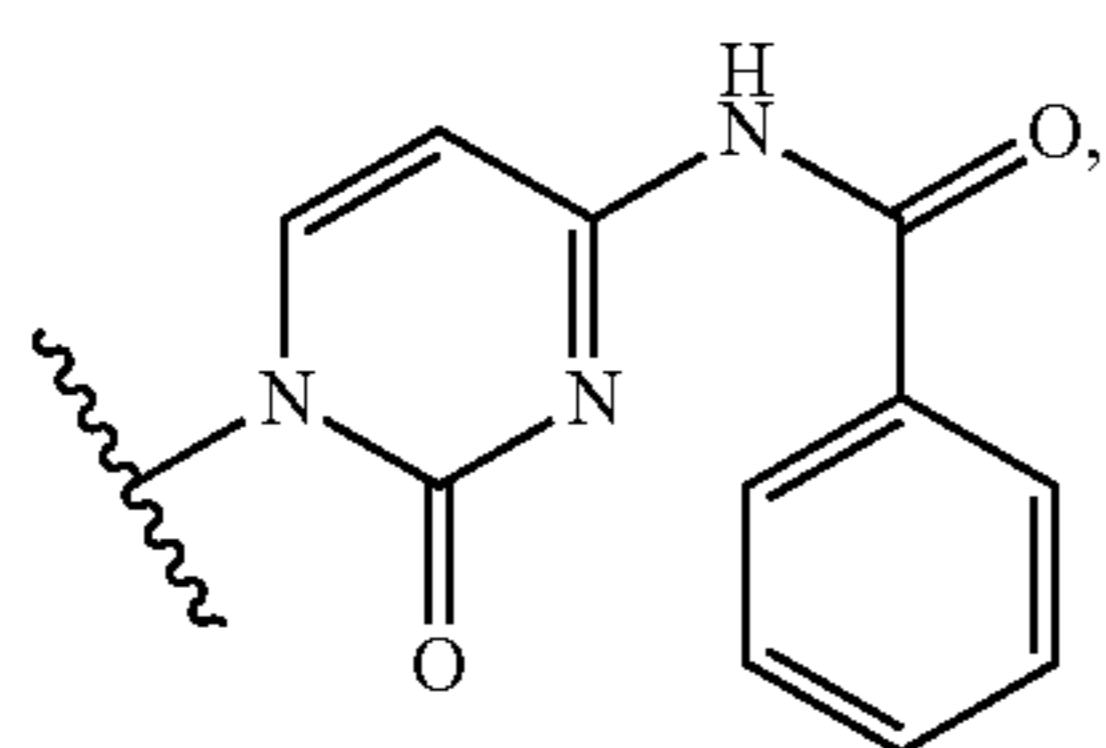
(A10a)

wherein:

n is an integer from 10 to 40,

R^1 is a support-medium, and

R^4 is, independently for each occurrence, selected from the group consisting of:



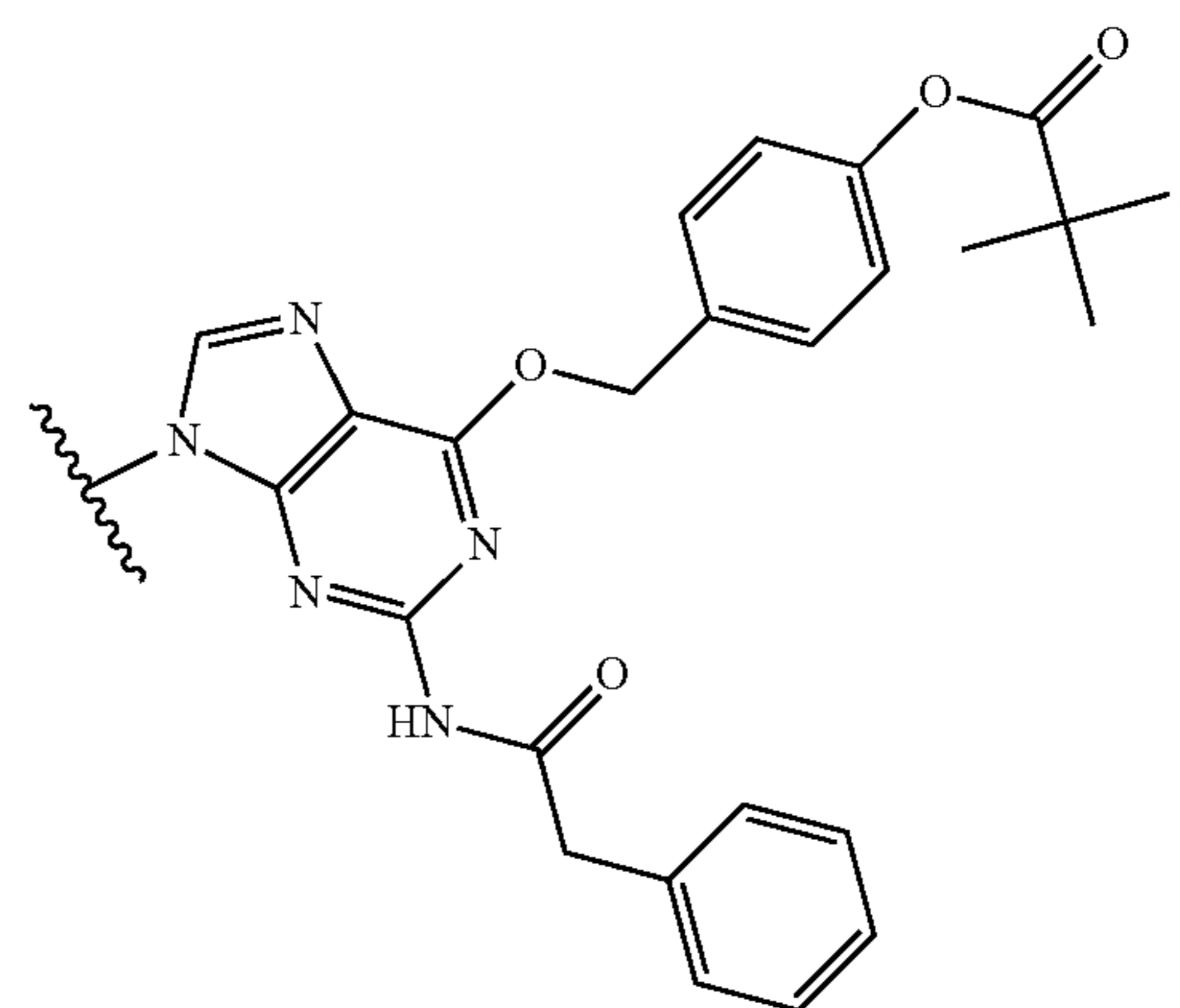
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(PC)

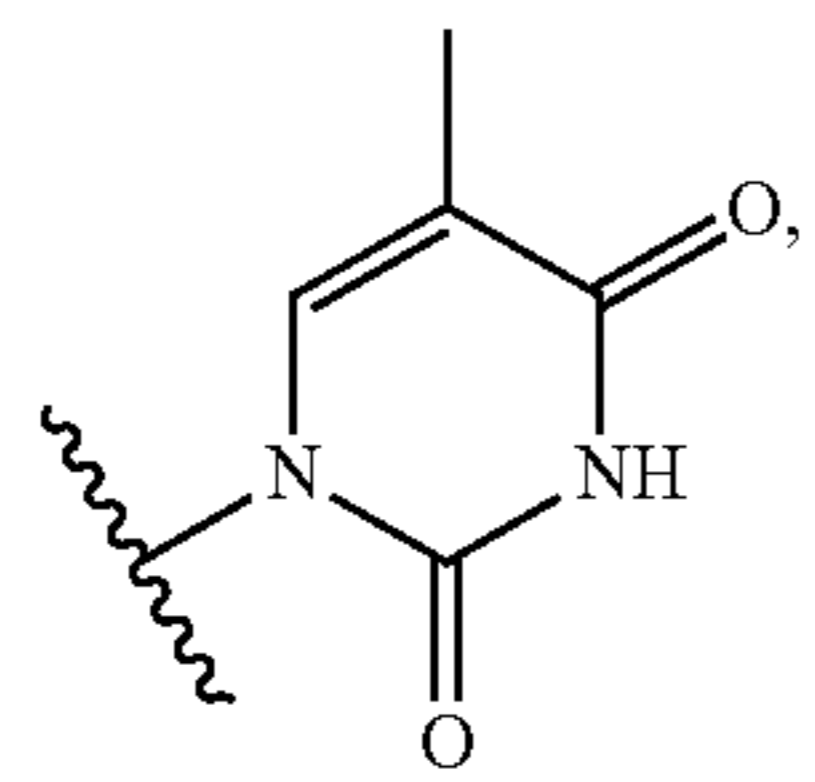
65



(DPG)

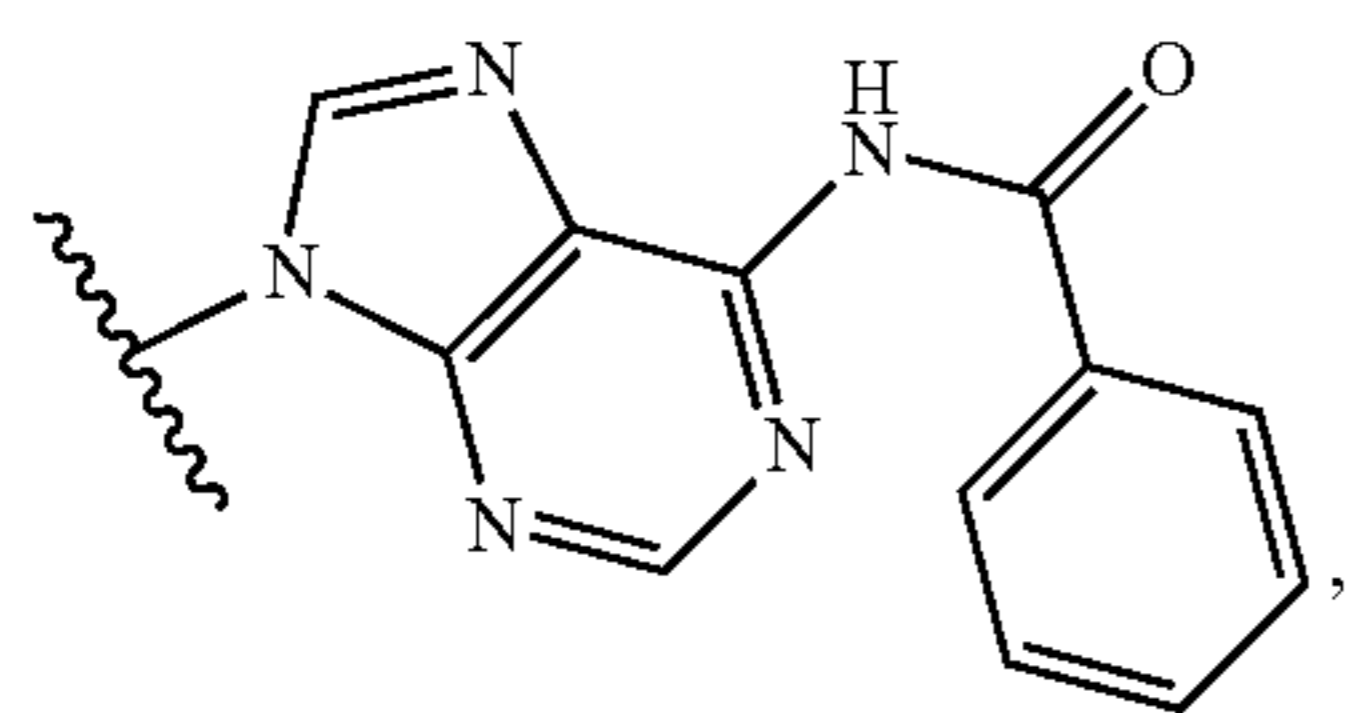
75

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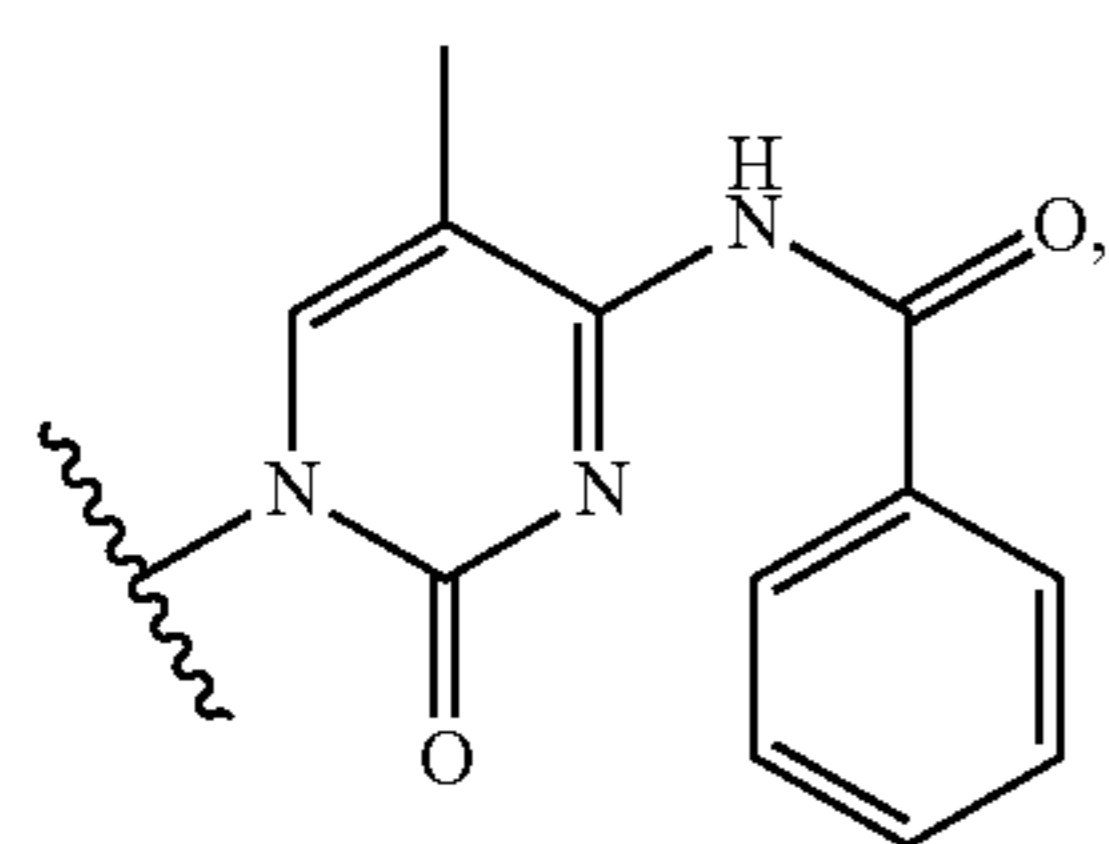


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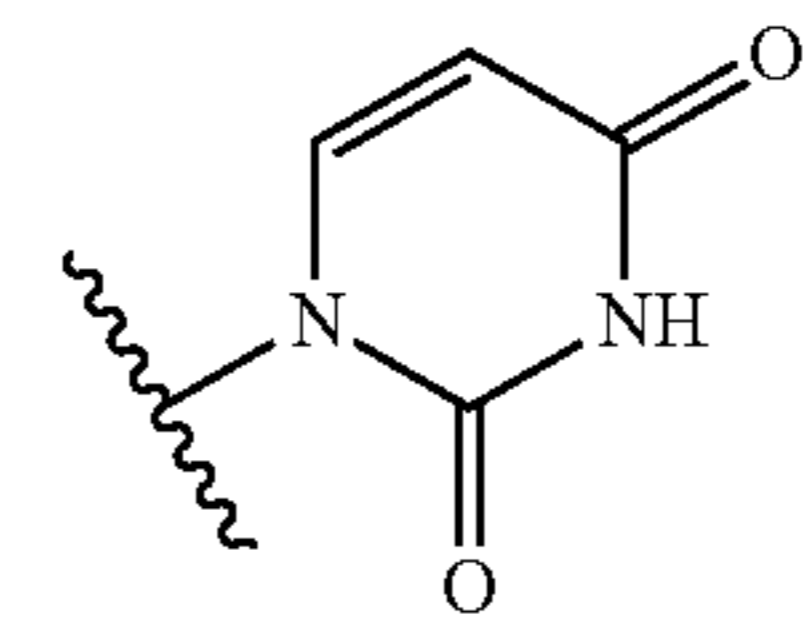
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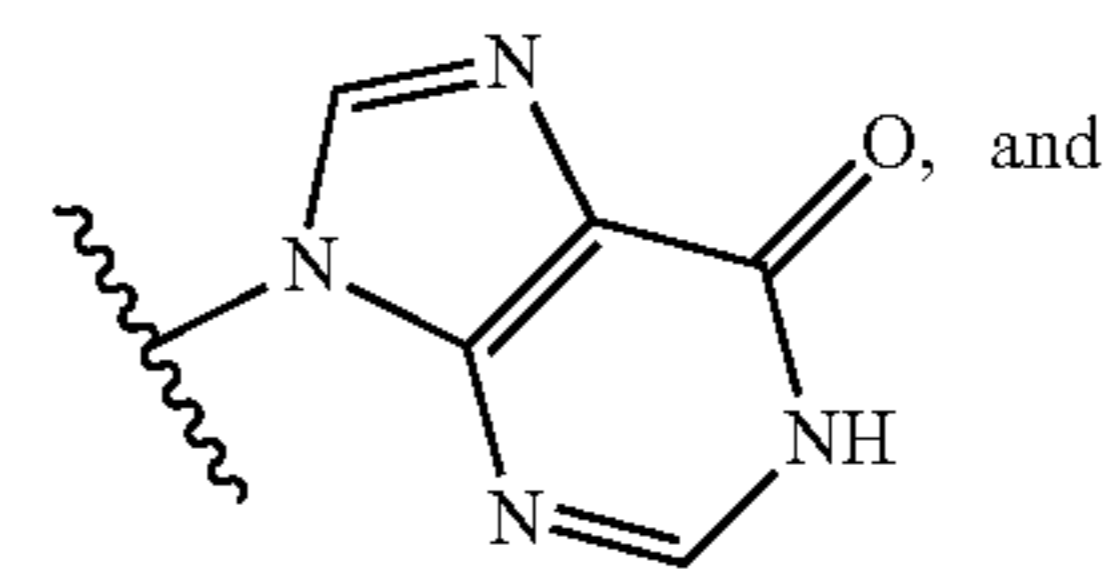
35

76

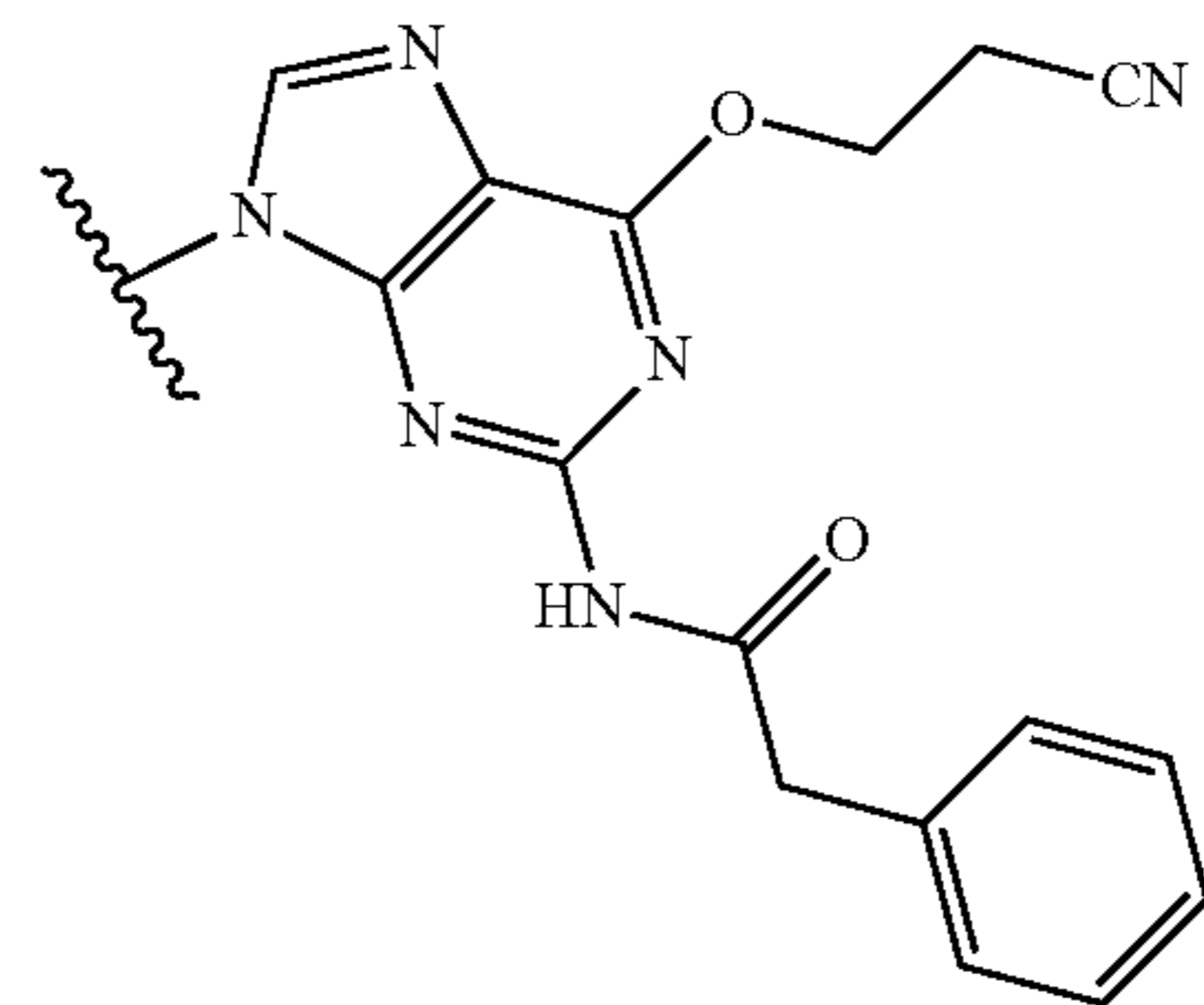
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(U)

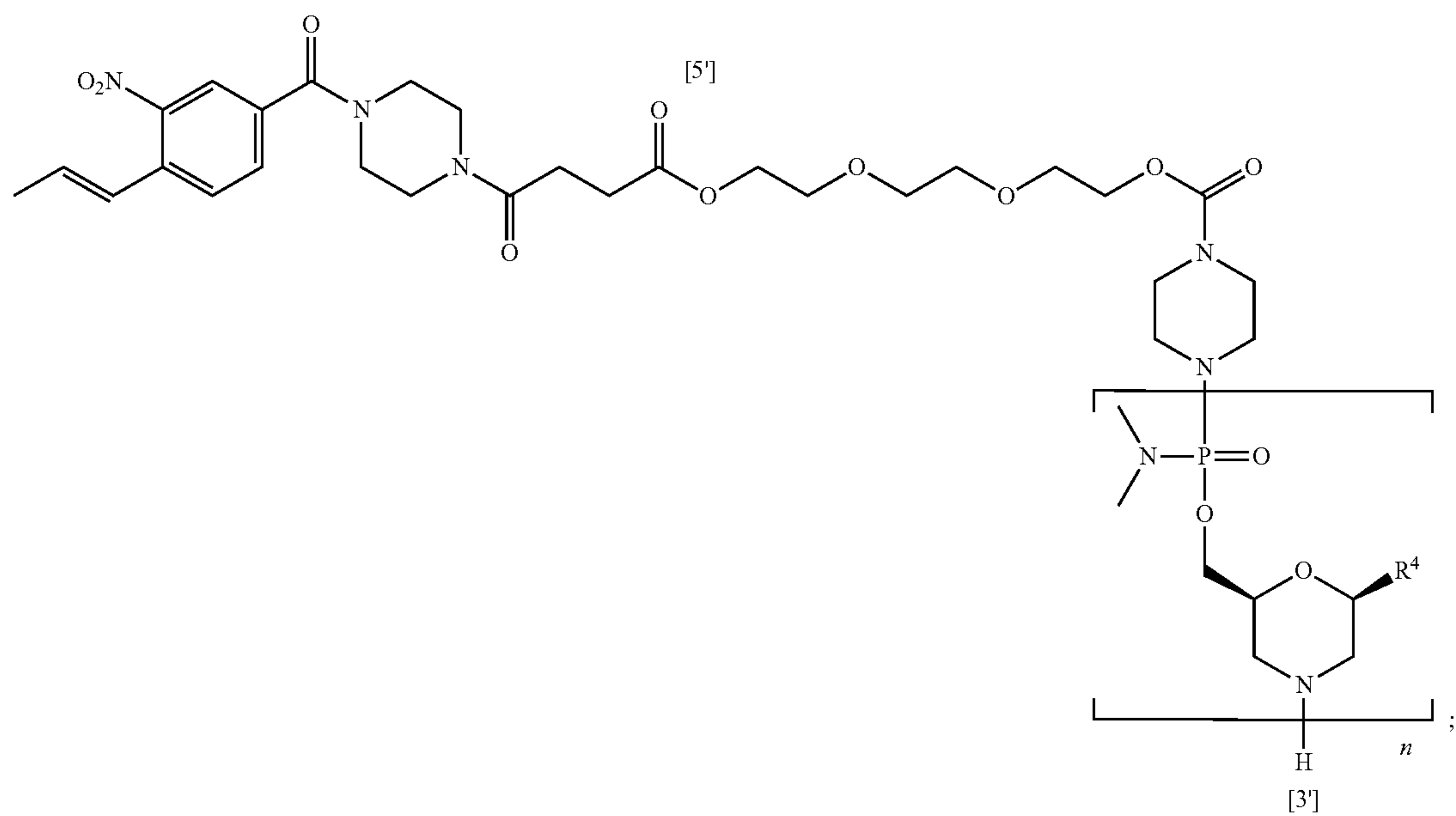


(I)



(PG)

In another embodiment, the compound of Formula (A11) is of Formula (A11a):



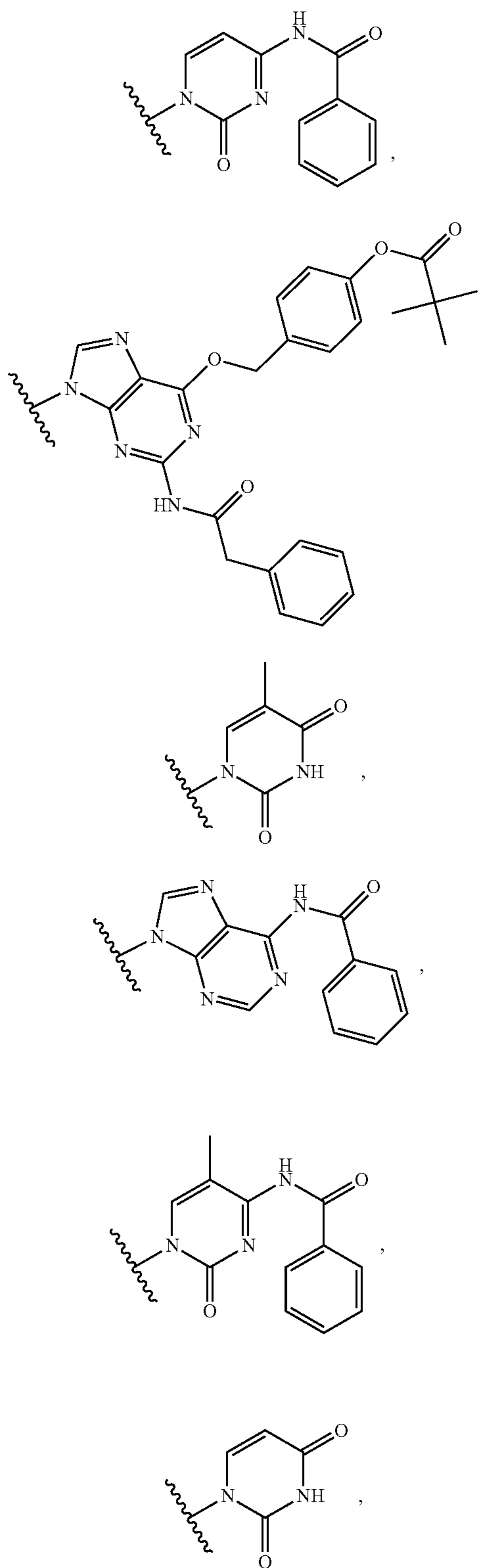
(A11a)

77

wherein:

n is an integer from 10 to 40, and

R⁴ is, independently for each occurrence, selected from the group consisting of:



78

-continued

(I)

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(PC)

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(DPG)

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(T)

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(PA)

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(P5mC)

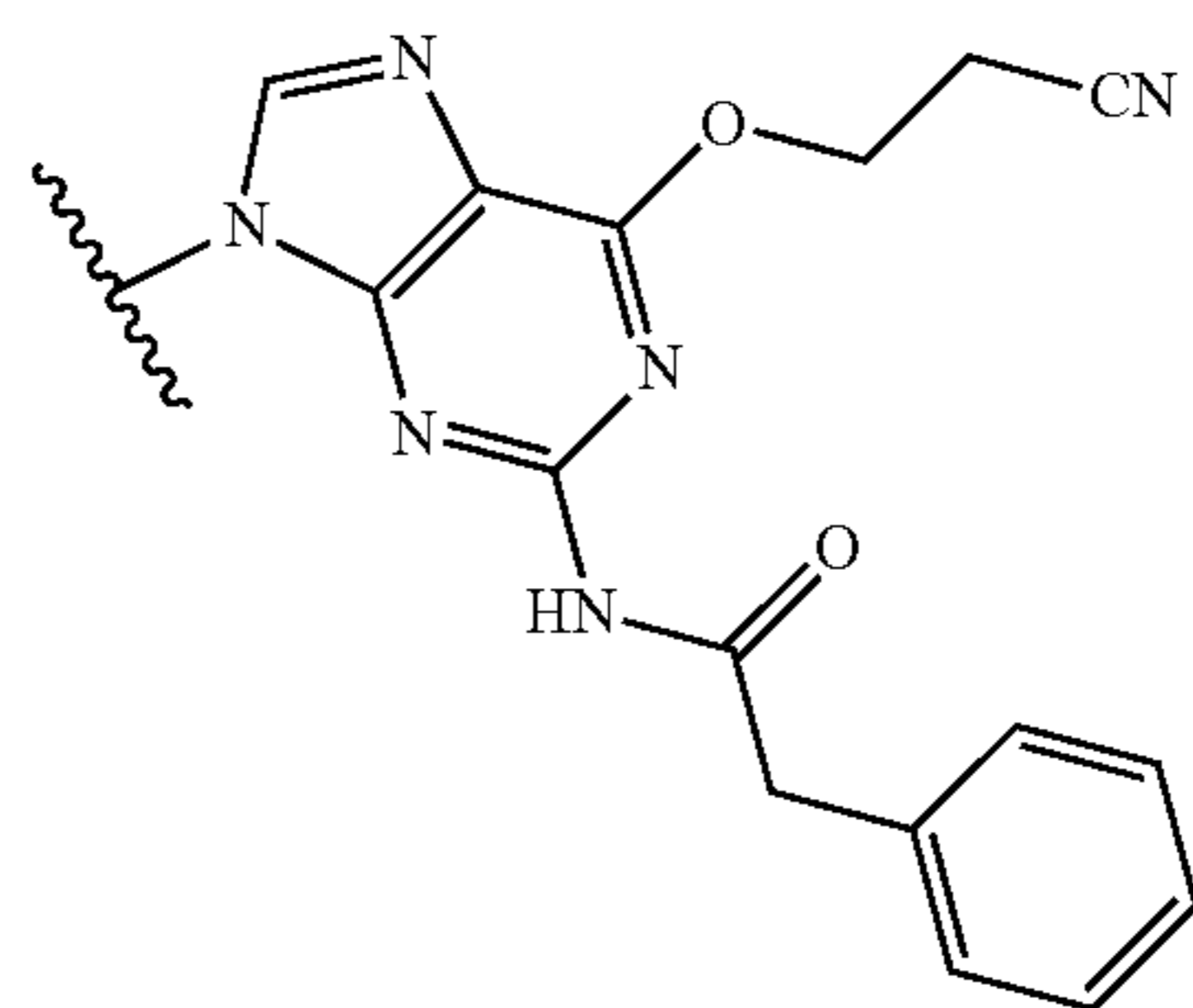
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(U)

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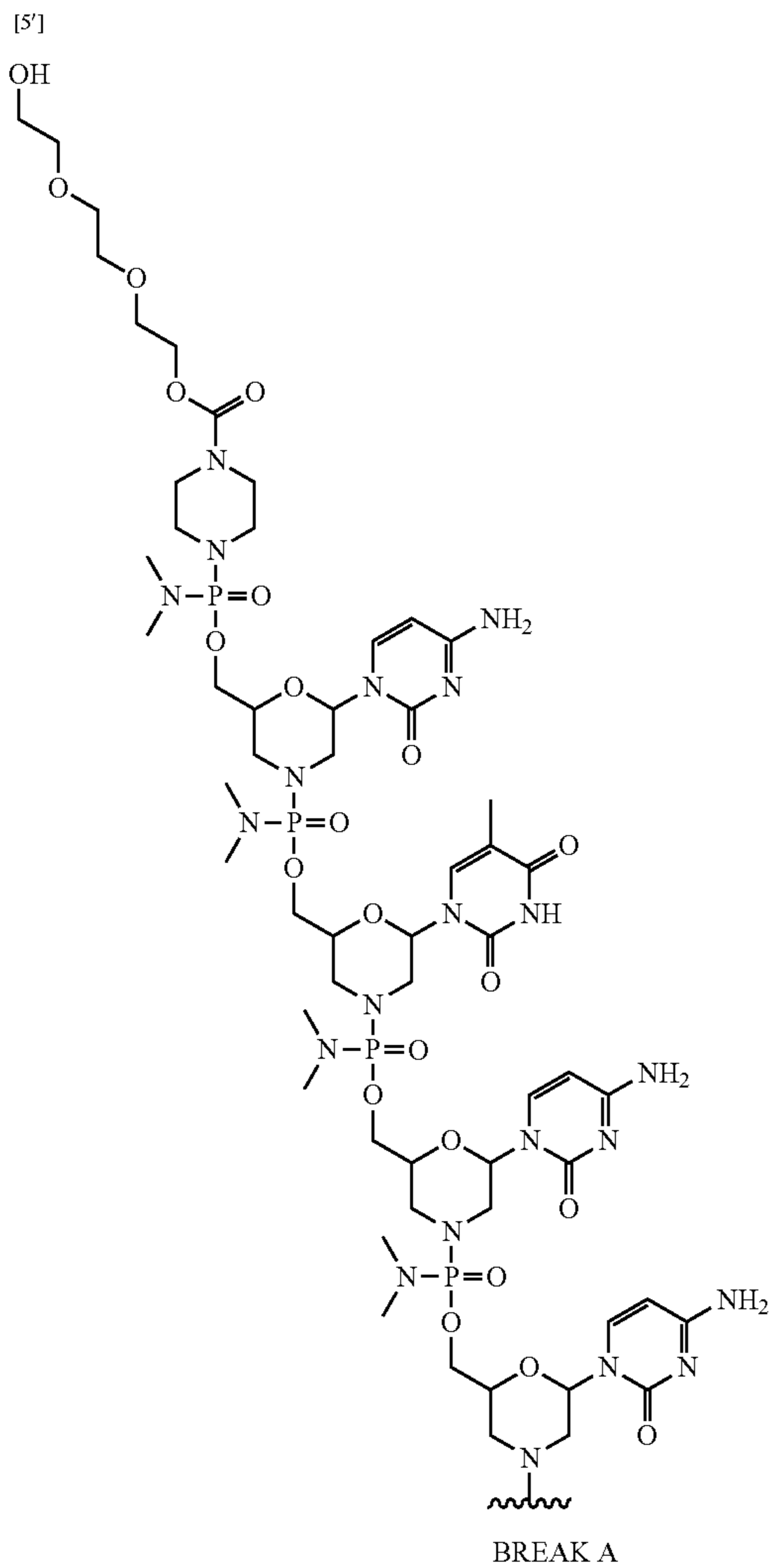


In an embodiment of the oligomeric compound of Formula (A), n is 30, and R² is at each position from 1 to 30 and 5' to 3':

Position No. 5' to 3'	R ²
1	C
2	T
3	C
4	C
5	A
6	A
7	C
8	A
9	T
10	C
11	A
12	A
13	G
14	G
15	A
16	A
17	G
18	A
19	T
20	G
21	G
22	C
23	A
24	T
25	T
26	T
27	C
28	T
29	A
30	G

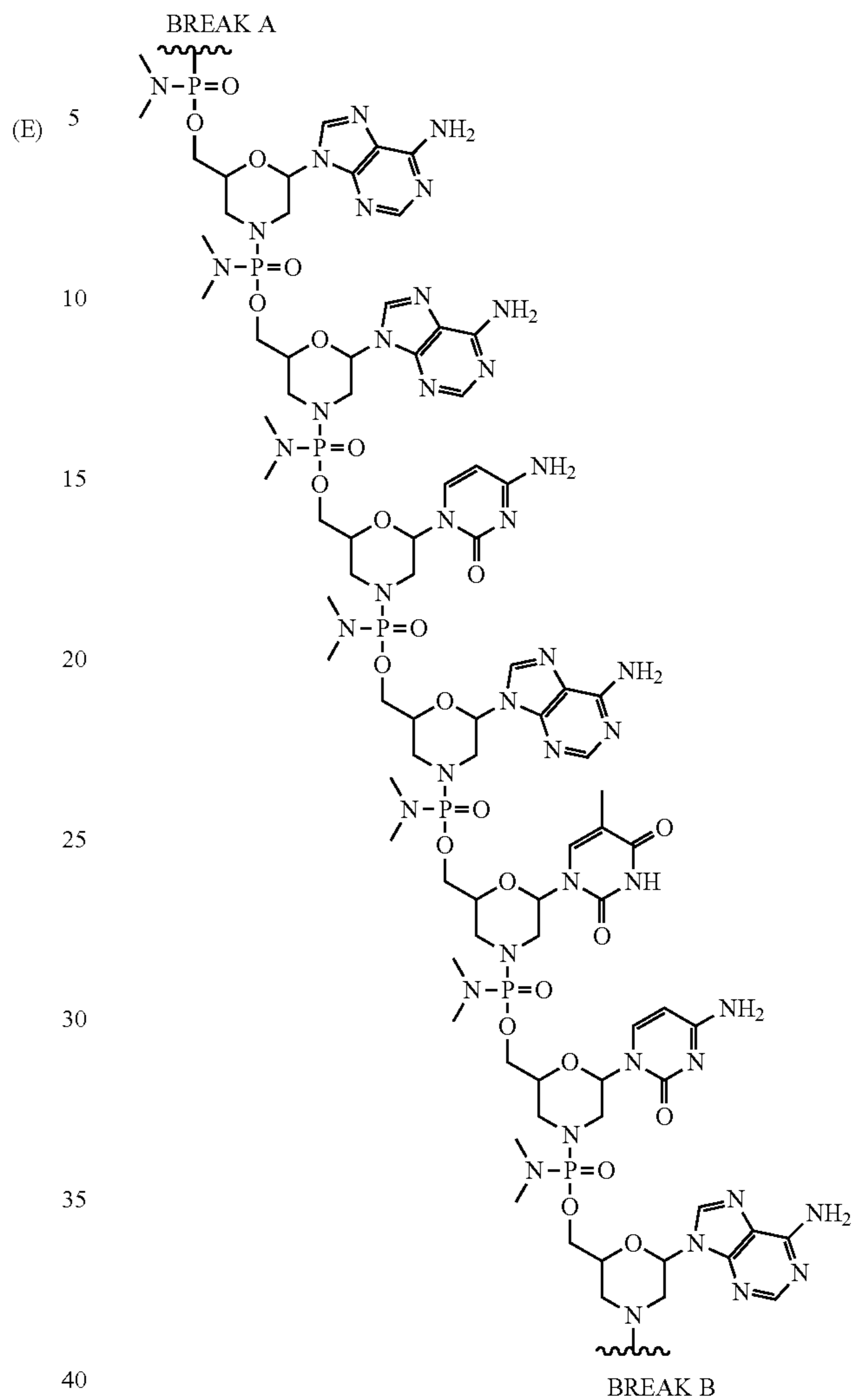
79

wherein the oligomeric compound of Formula (A) is a compound of Formula (E):



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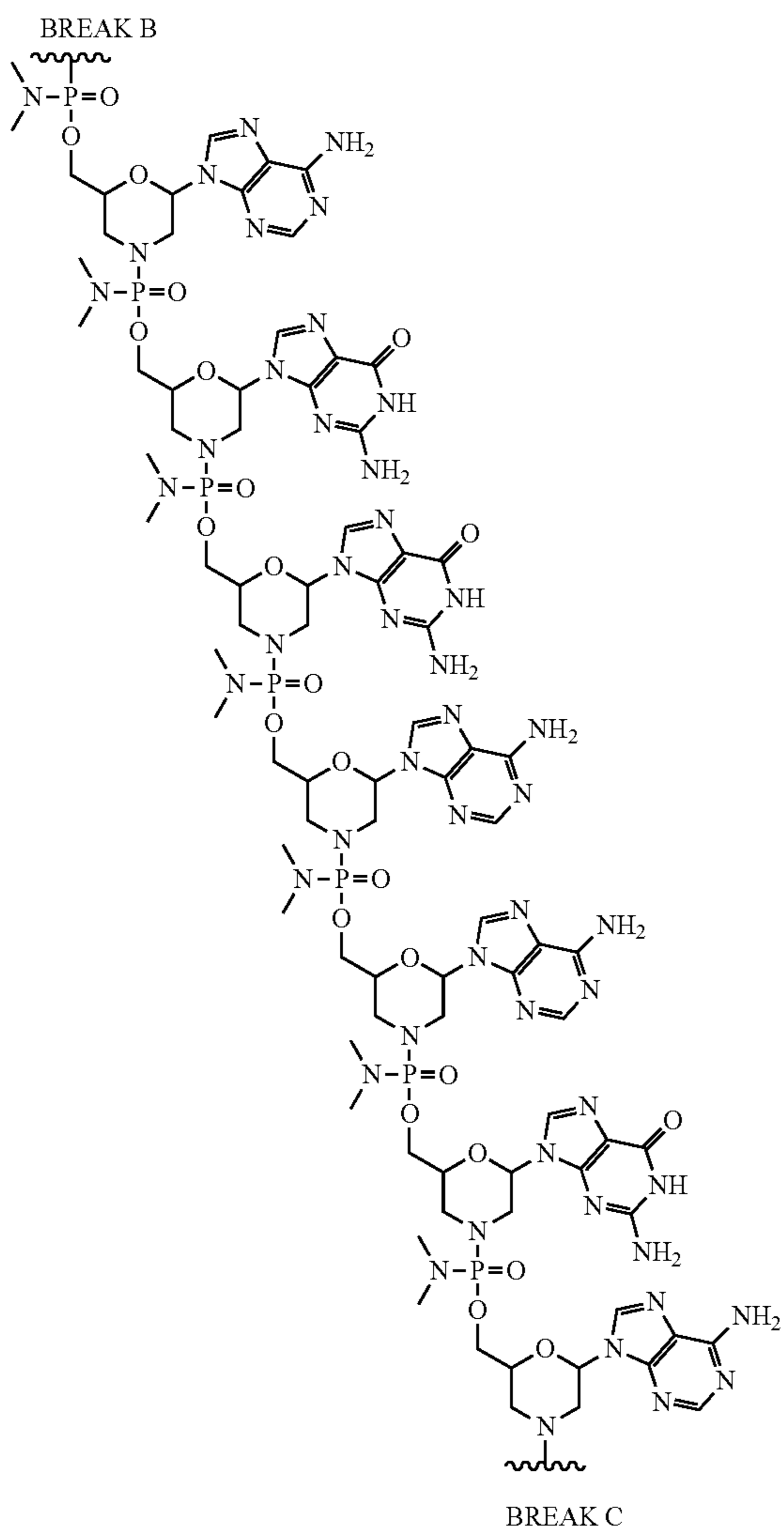
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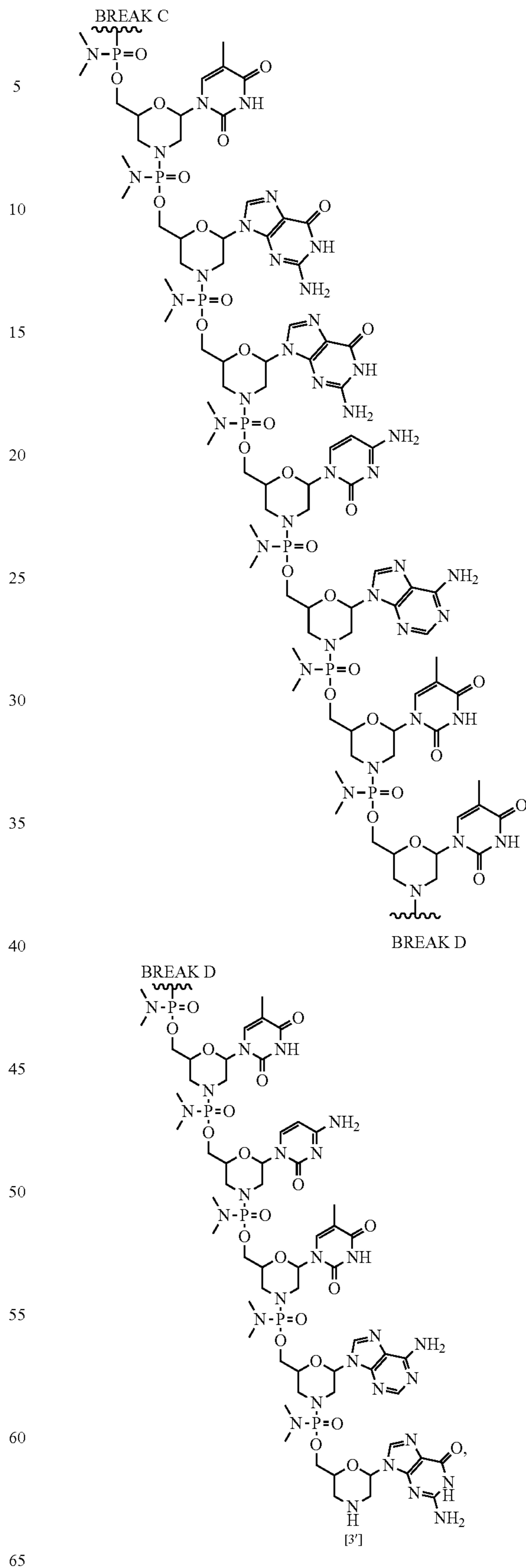
81

-continued



82

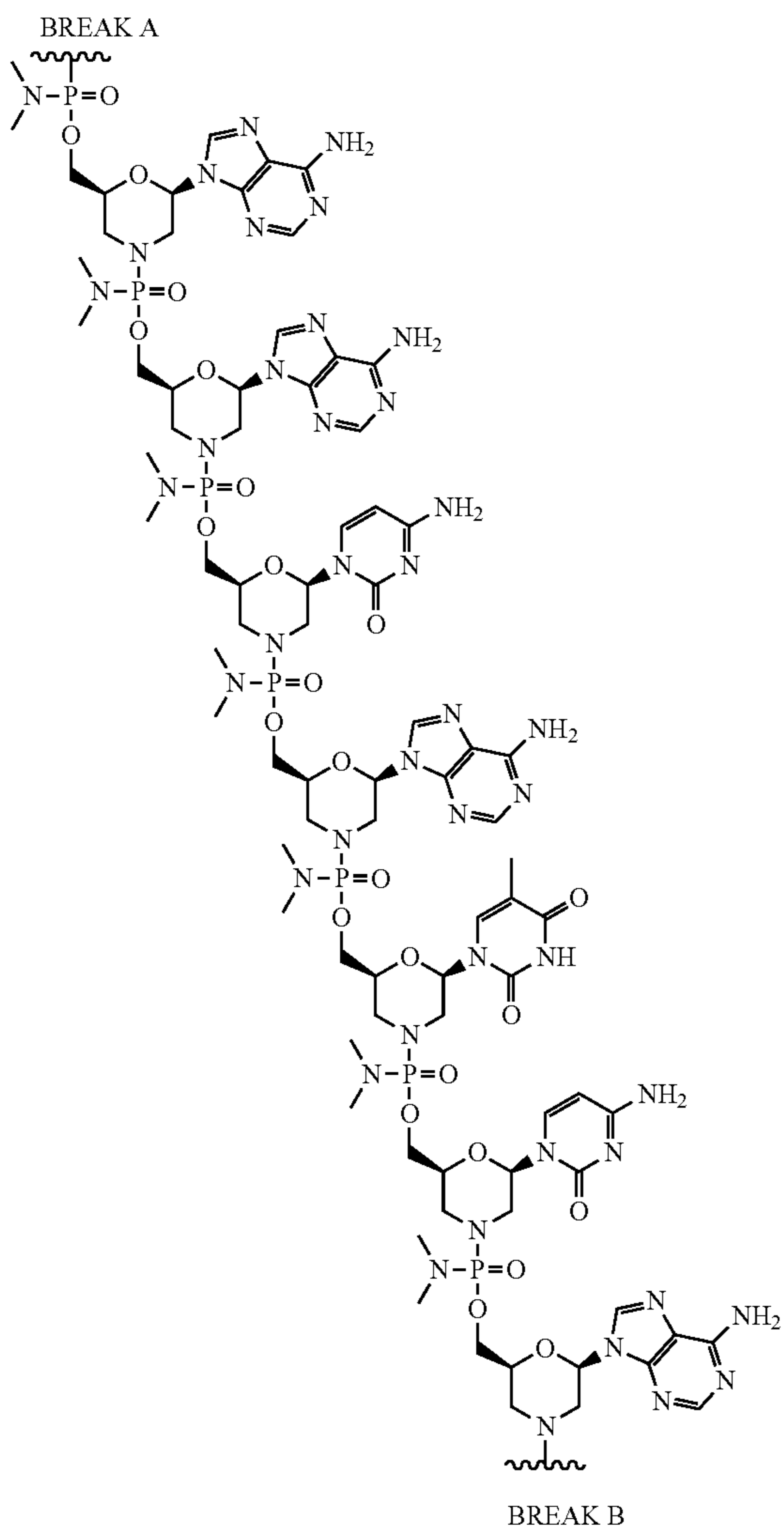
-continued



or a pharmaceutically acceptable salt thereof.

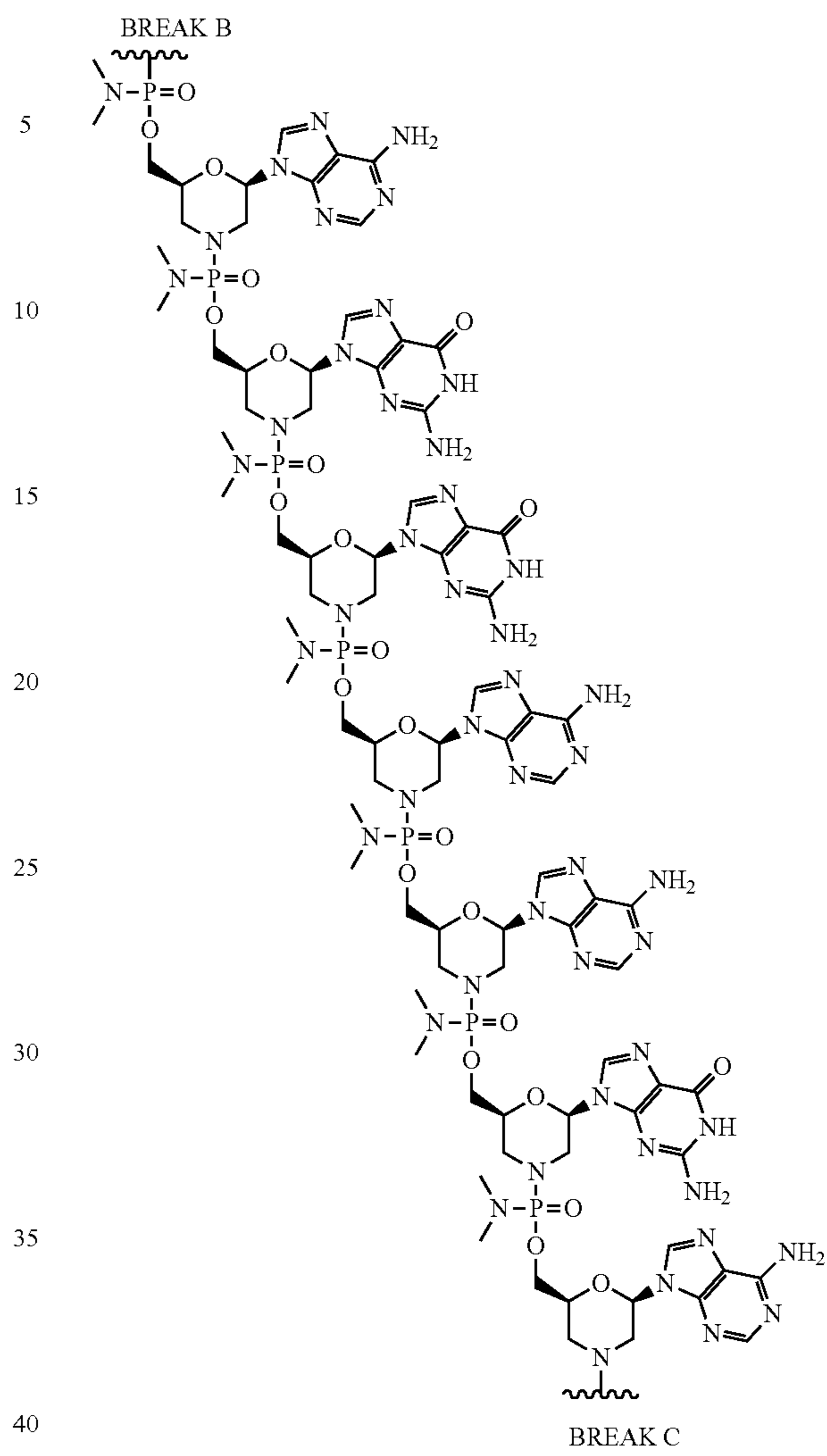
85

-continued



86

-continued



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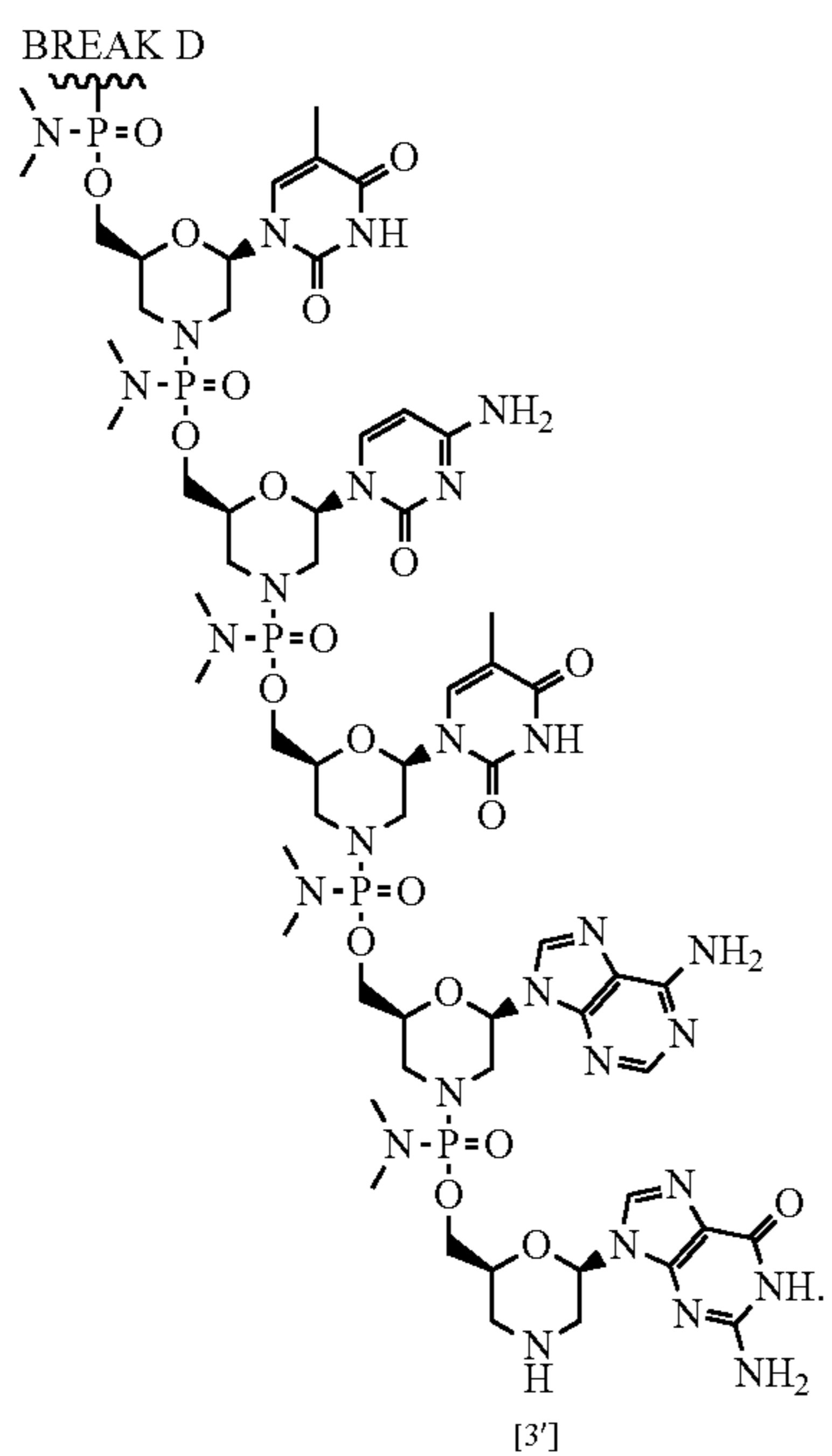
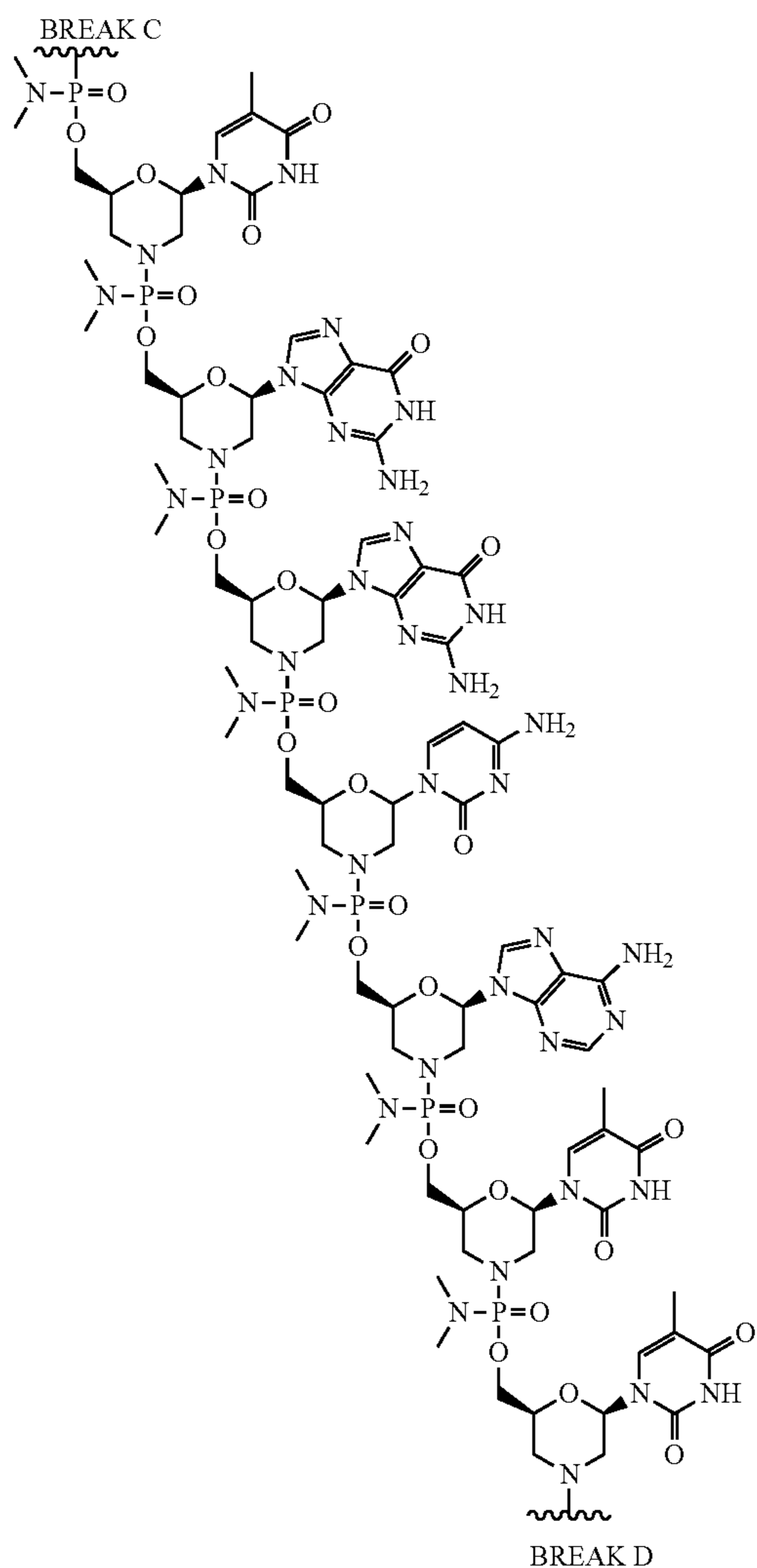
55

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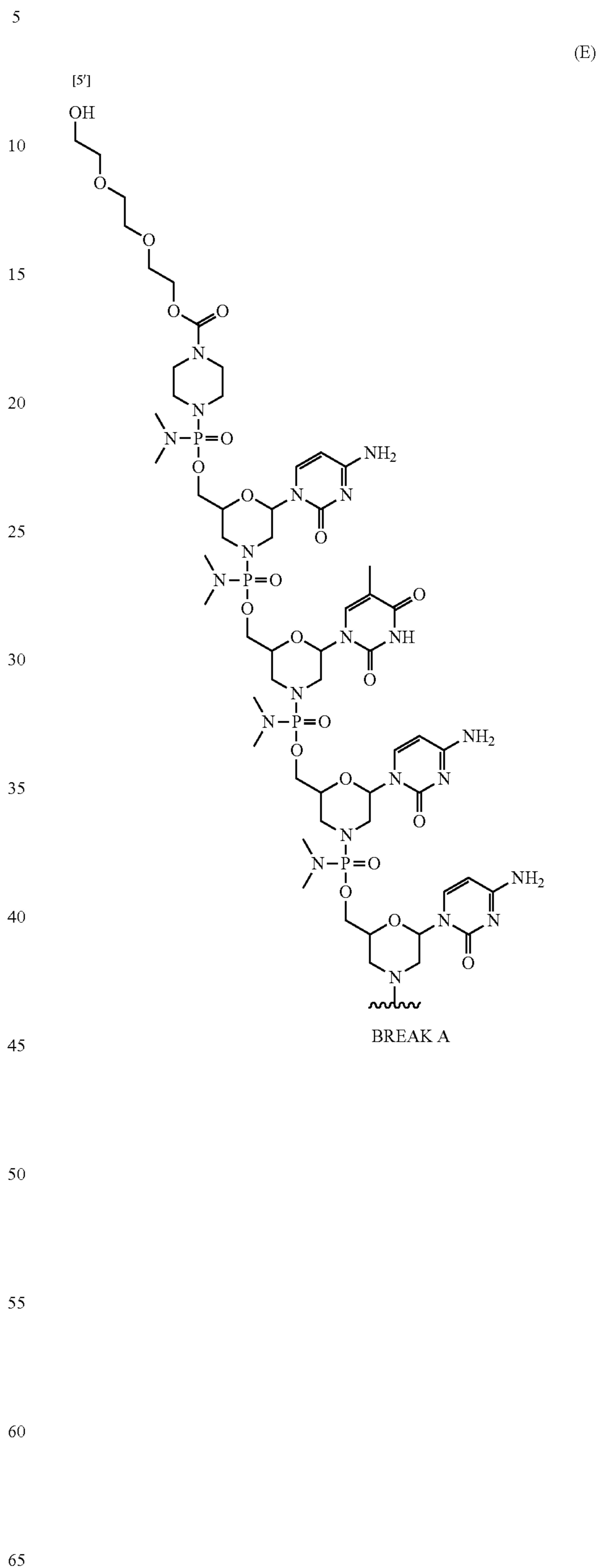
87

-continued



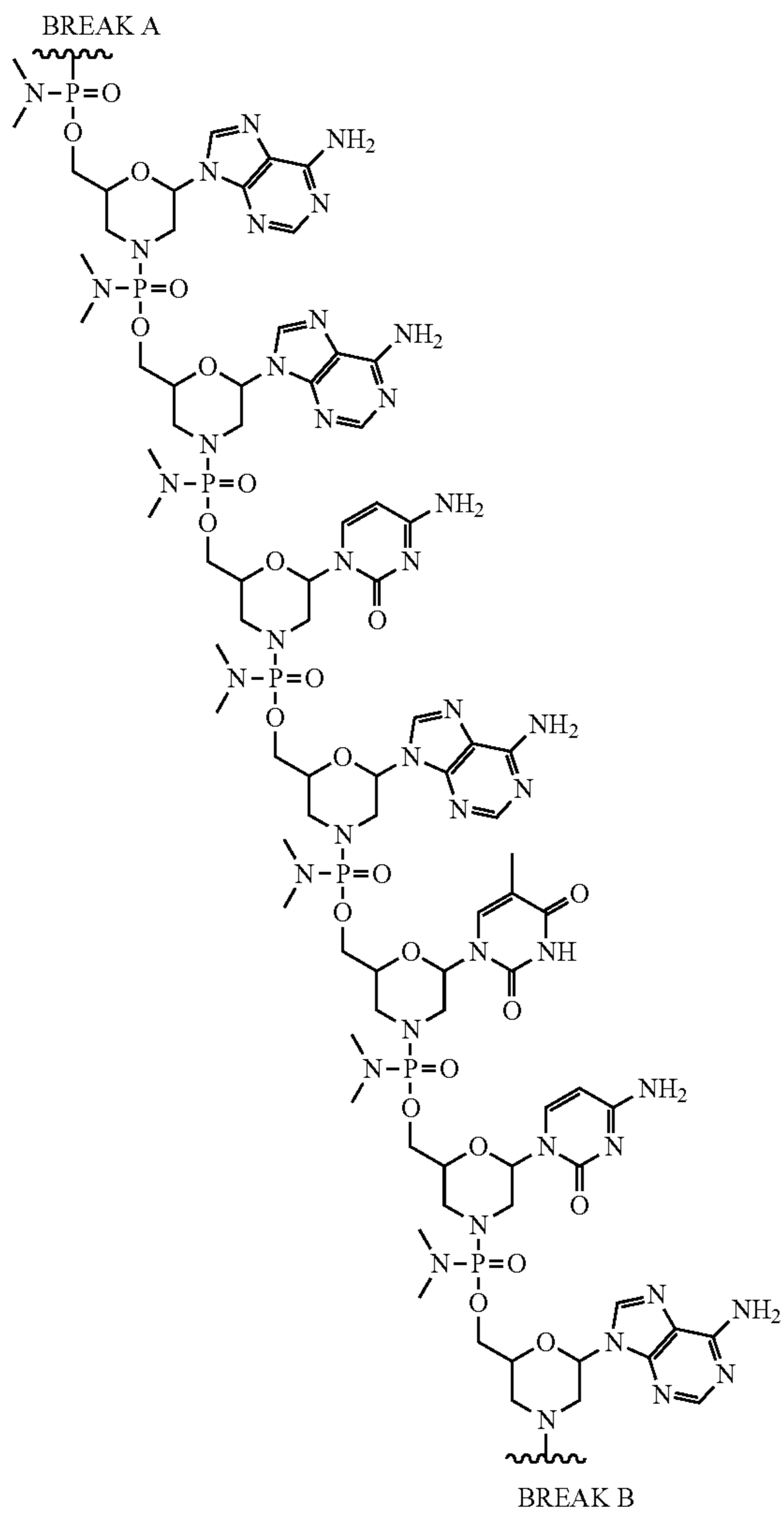
88

Thus, in one embodiment of the process described above, the oligomeric compound of Formula (A) is a compound of Formula (E):



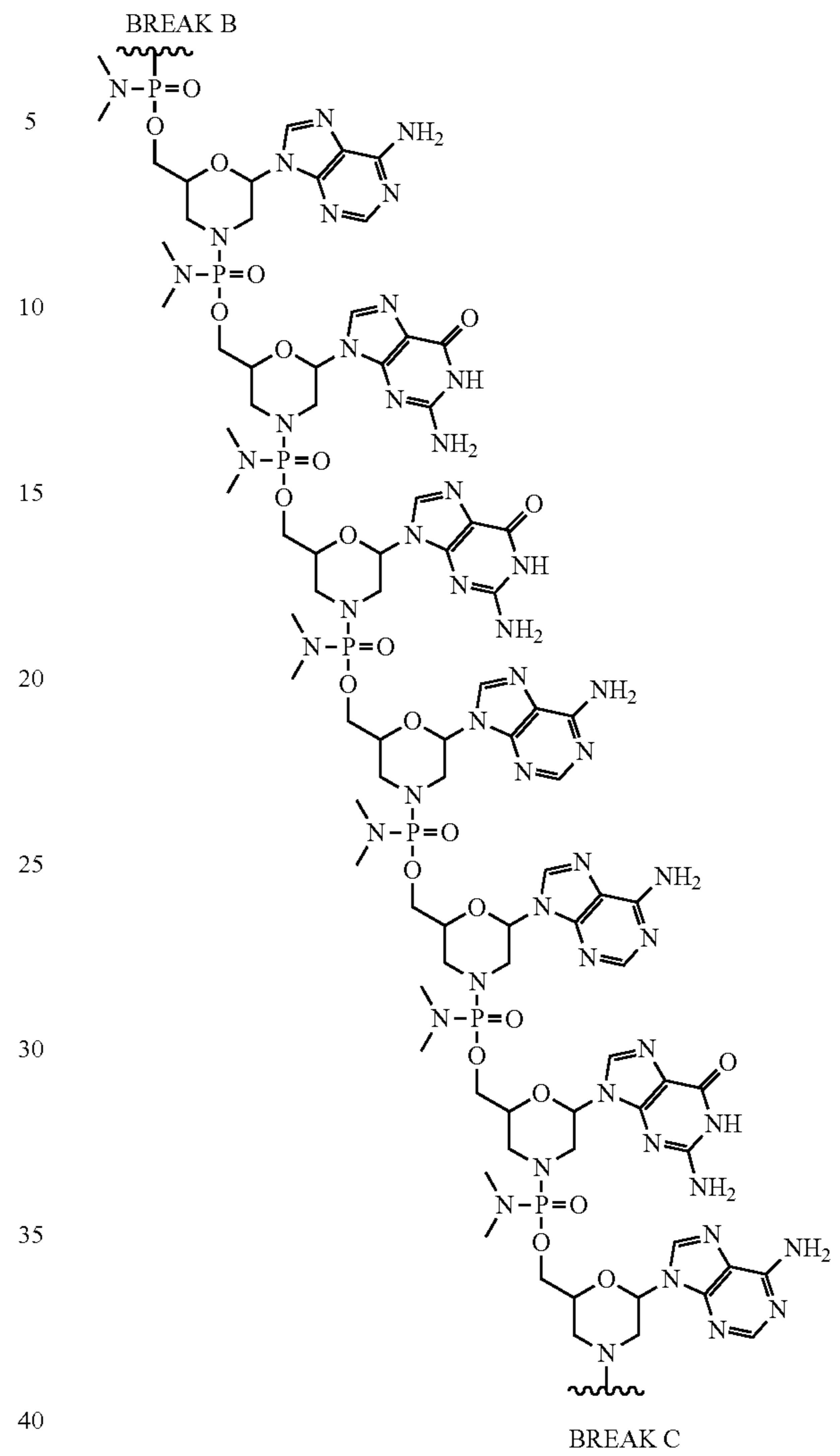
89

-continued



90

-continued



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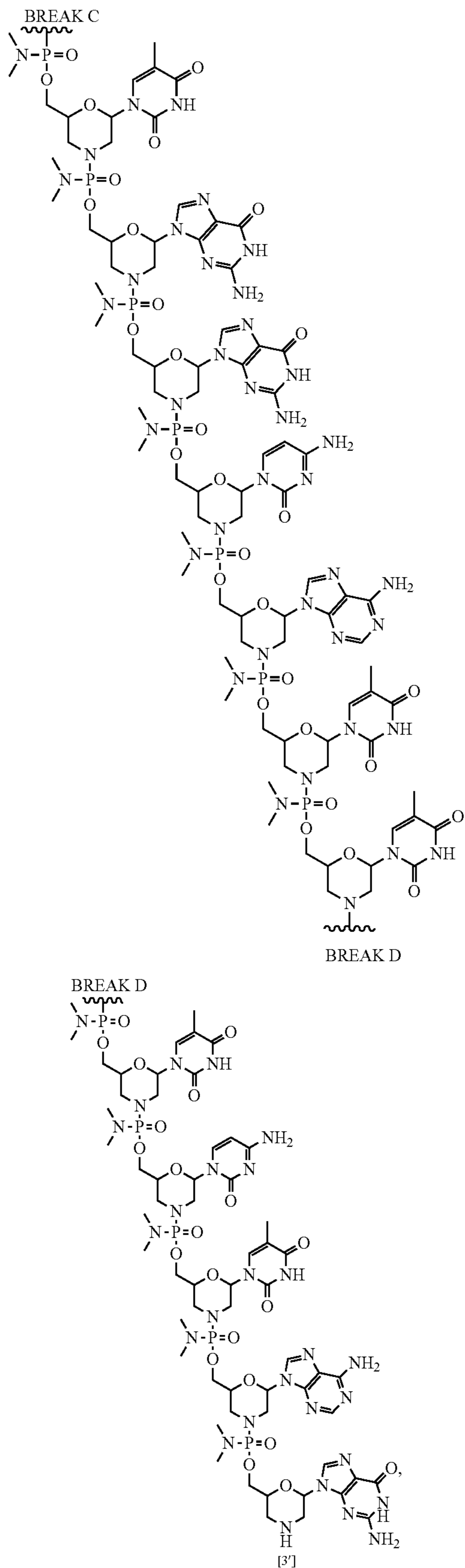
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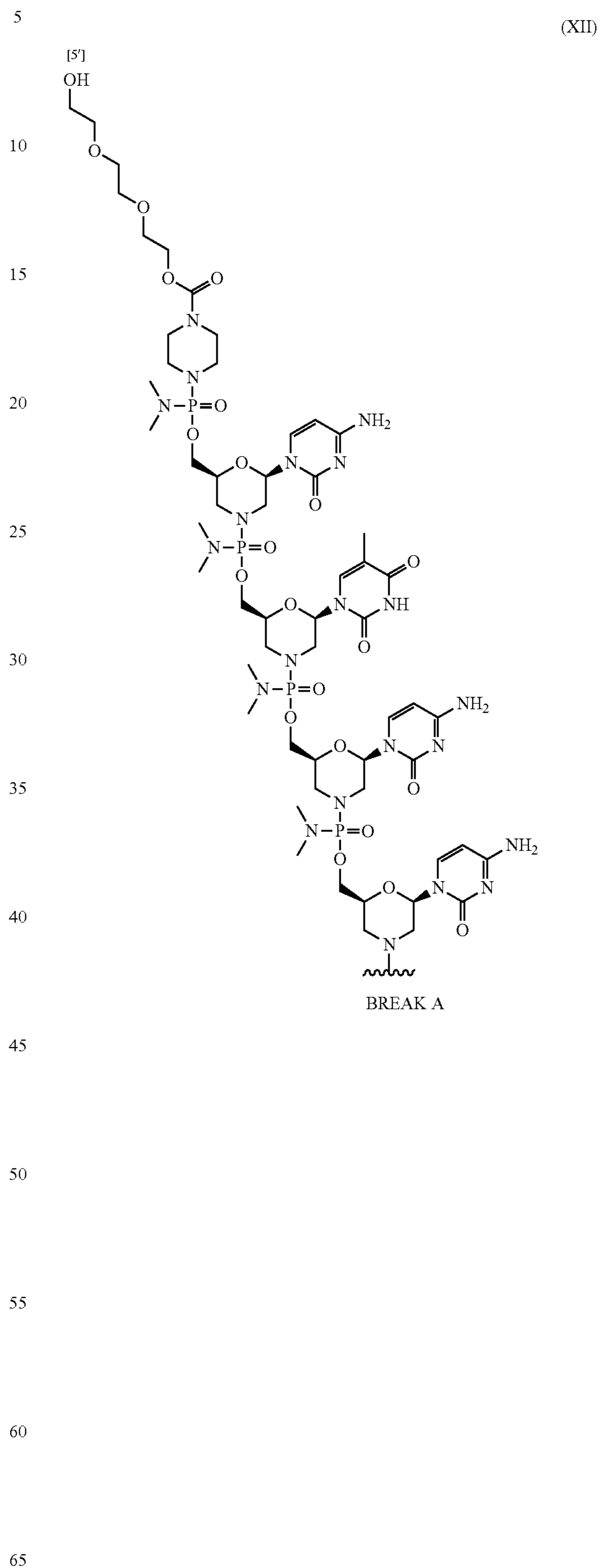
91

-continued



92

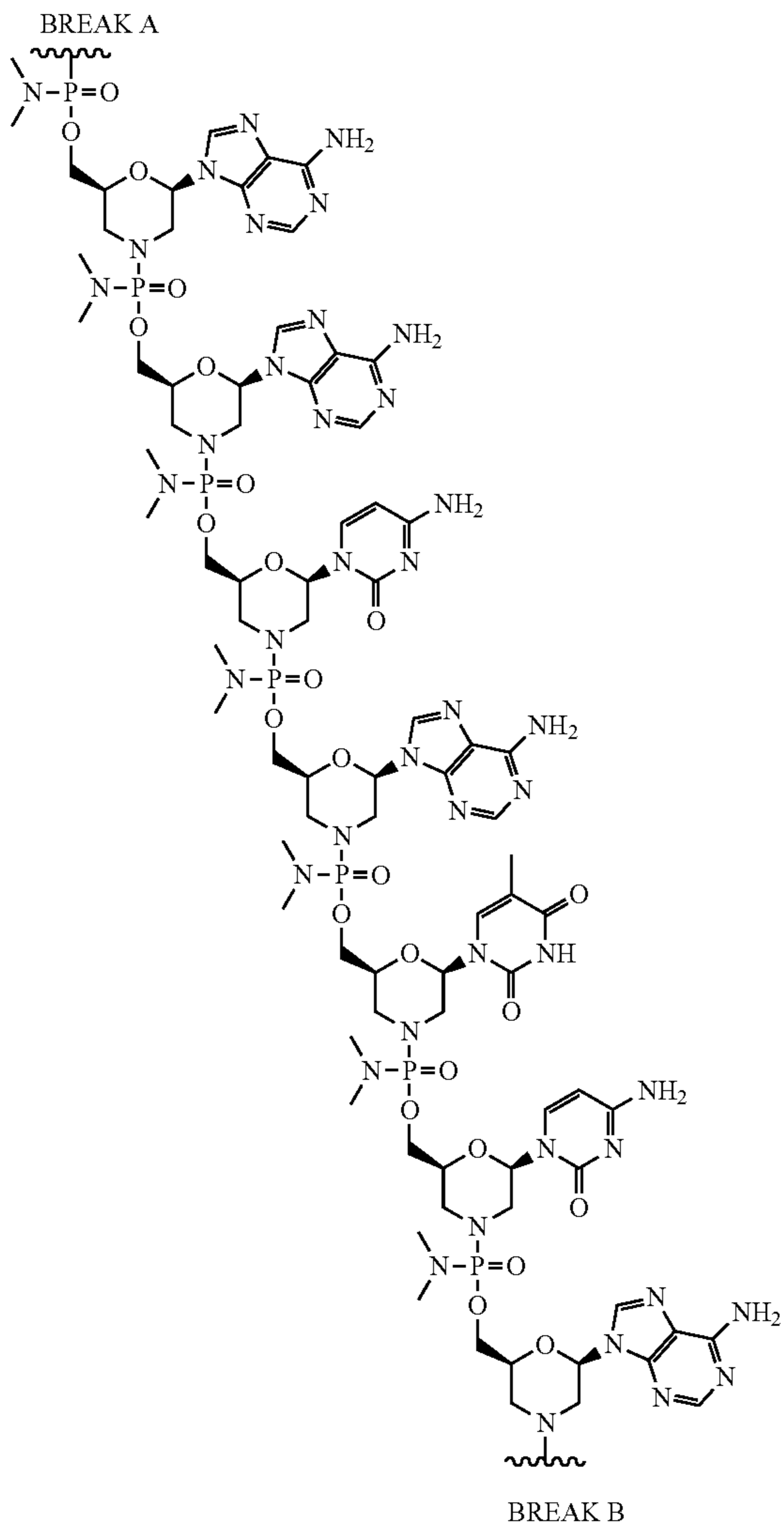
In yet another embodiment, the oligomeric compound of Formula (E) is an oligomeric compound of Formula (XII):



or a pharmaceutically acceptable salt thereof.

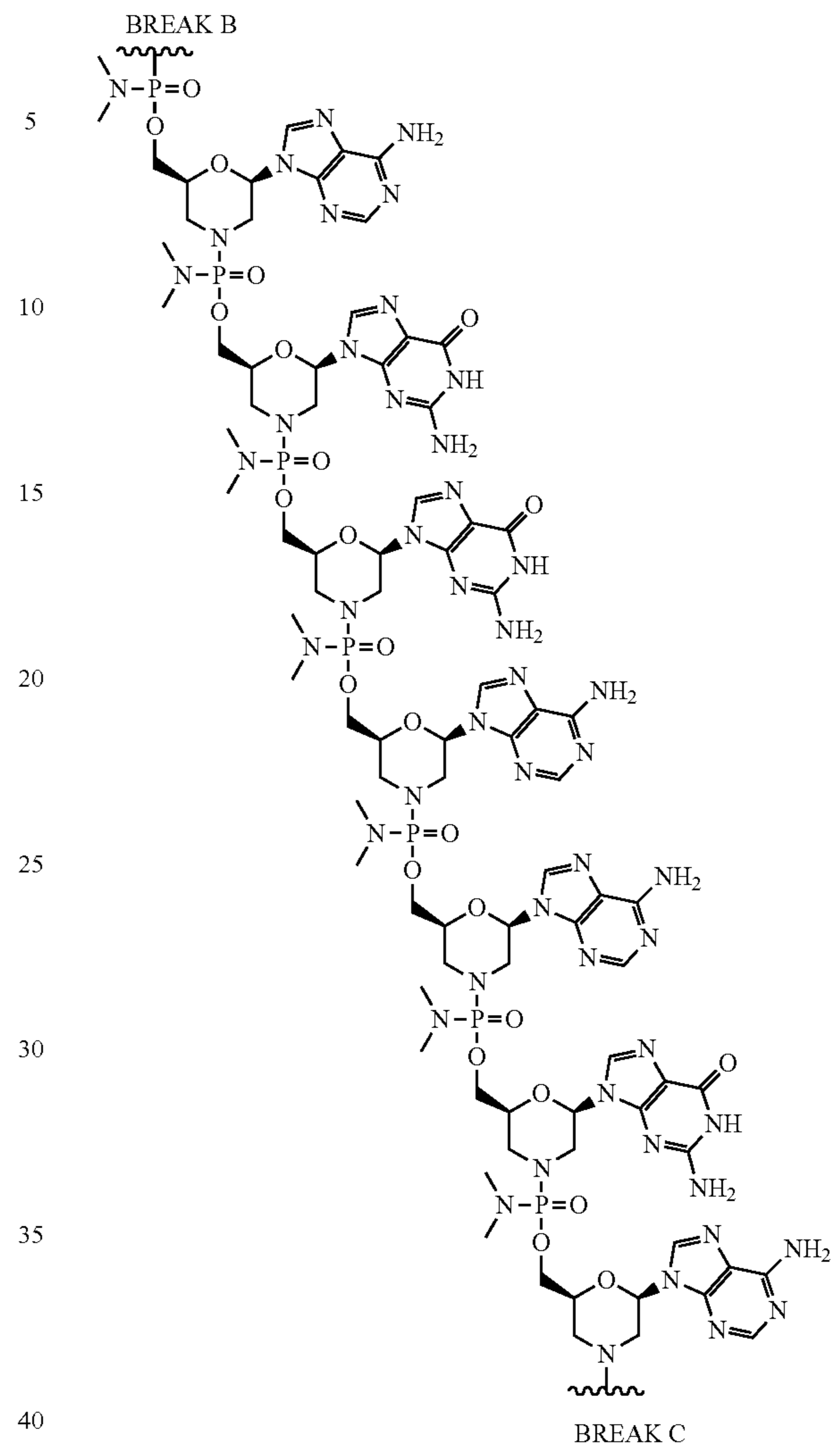
93

-continued



94

-continued



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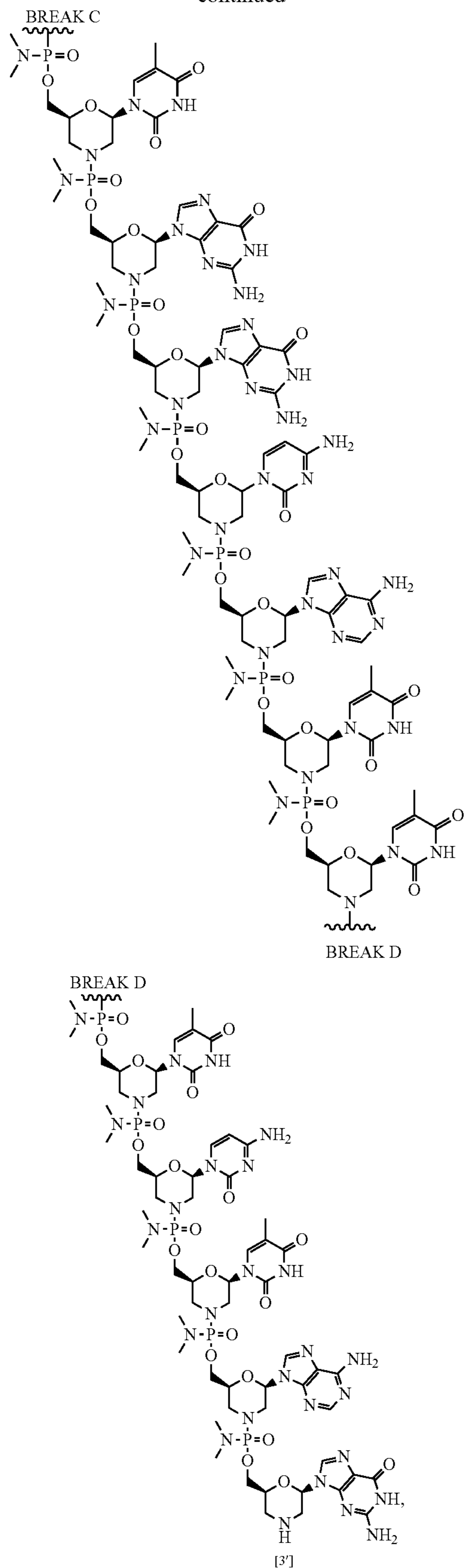
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95

-continued

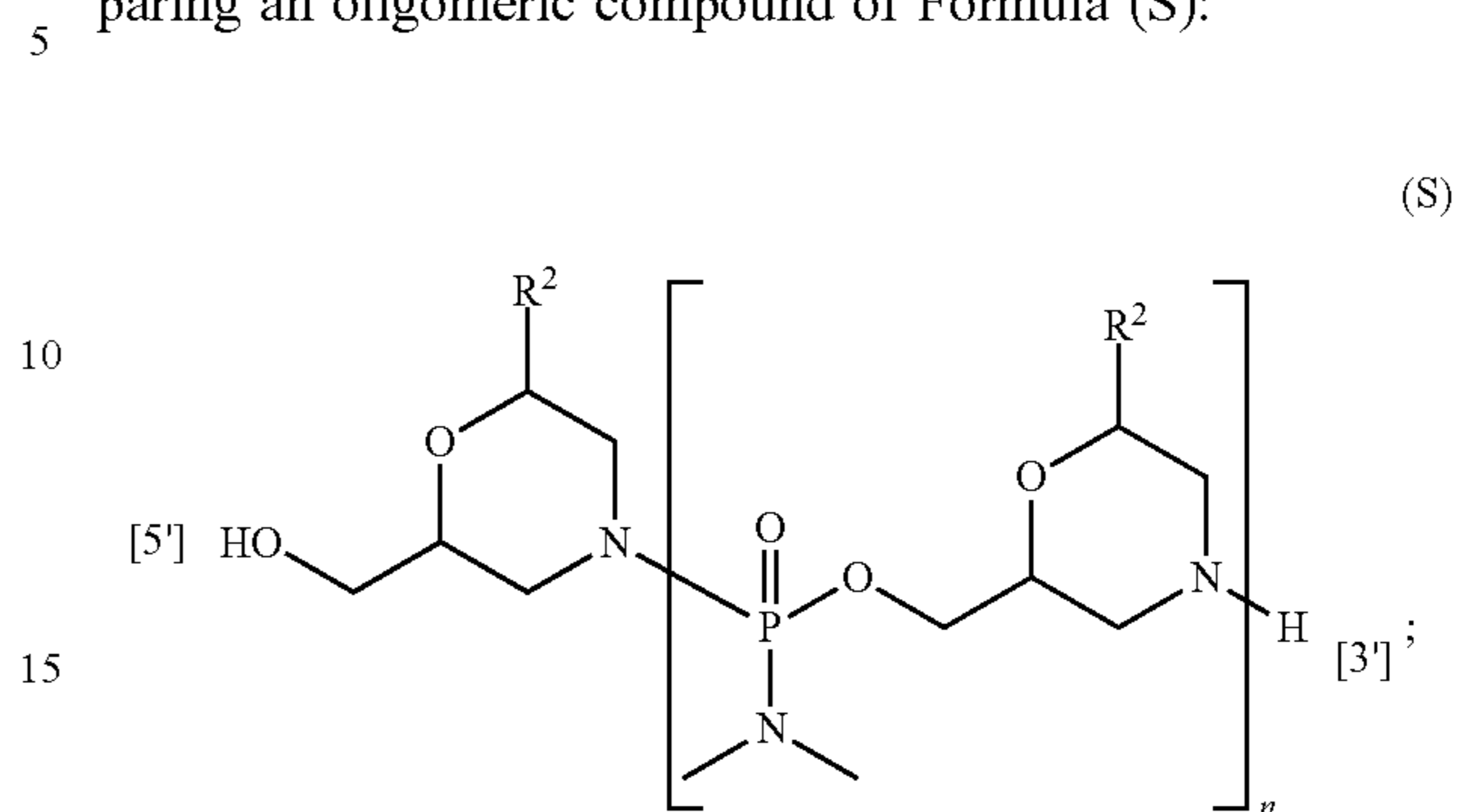


or a pharmaceutically acceptable salt thereof.

96

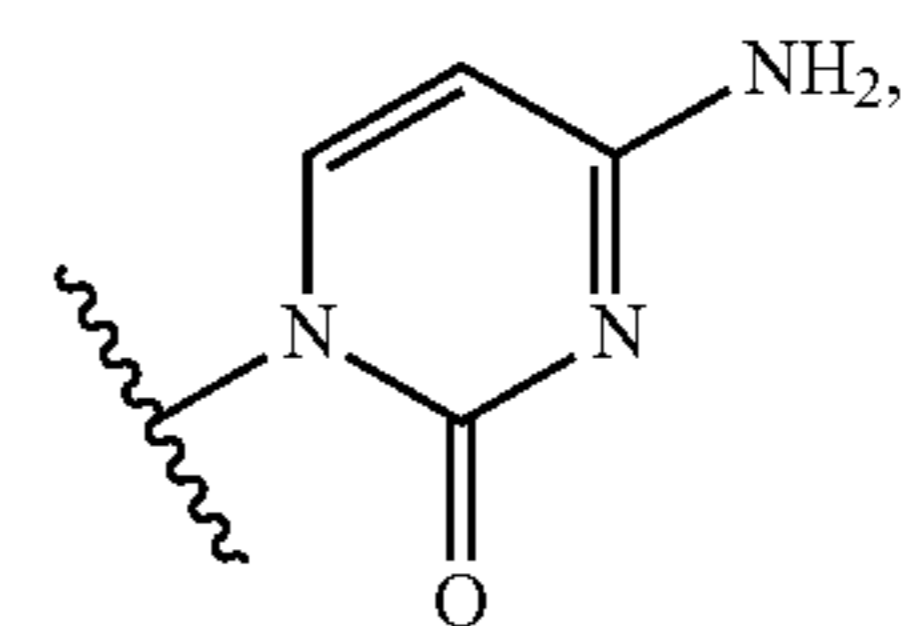
In still another embodiment, R³ is, at each occurrence, trityl.

In another aspect, provided herein is a process for preparing an oligomeric compound of Formula (S):

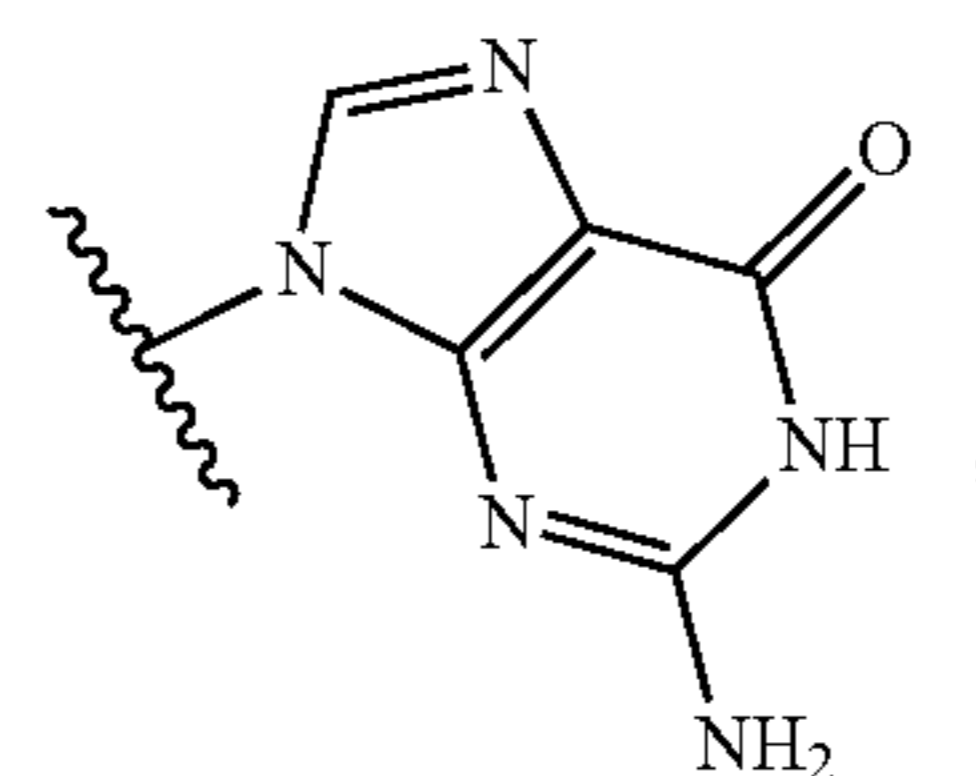


20 wherein n is an integer from 10 to 40, and each R² is, independently for each occurrence, selected from the group consisting of:

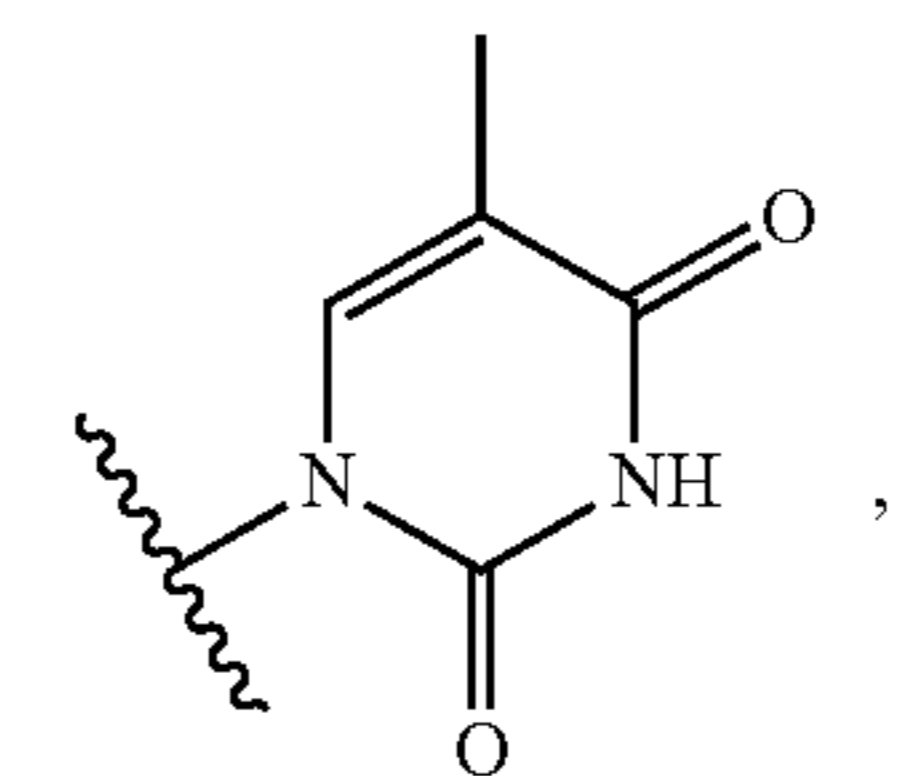
25 (C)



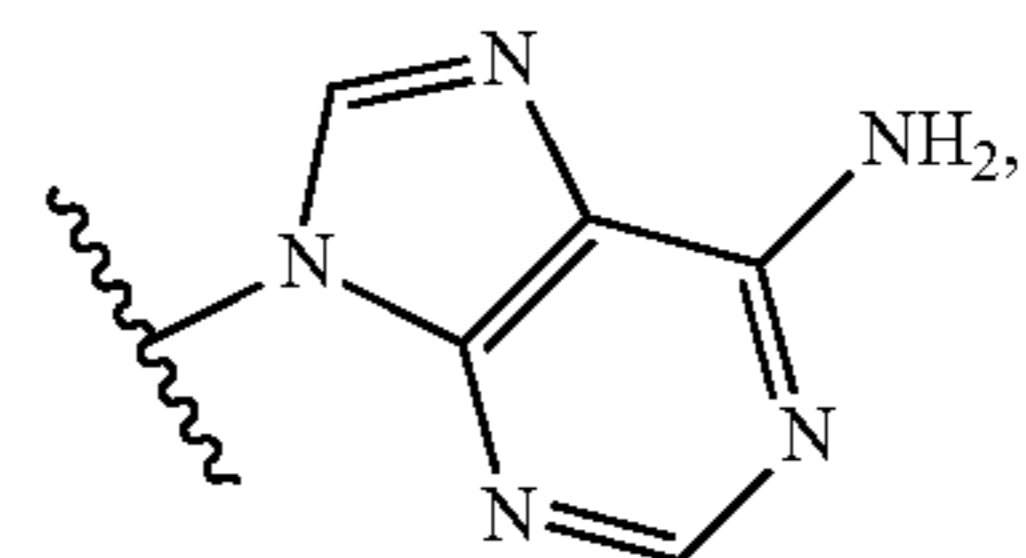
30 (G)



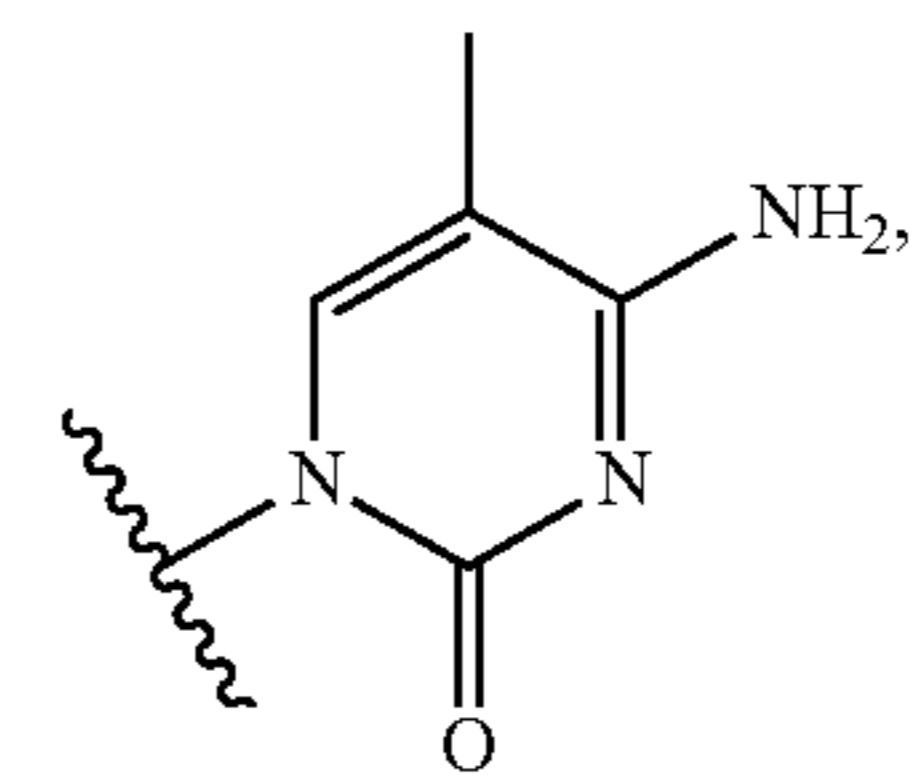
35 (T)



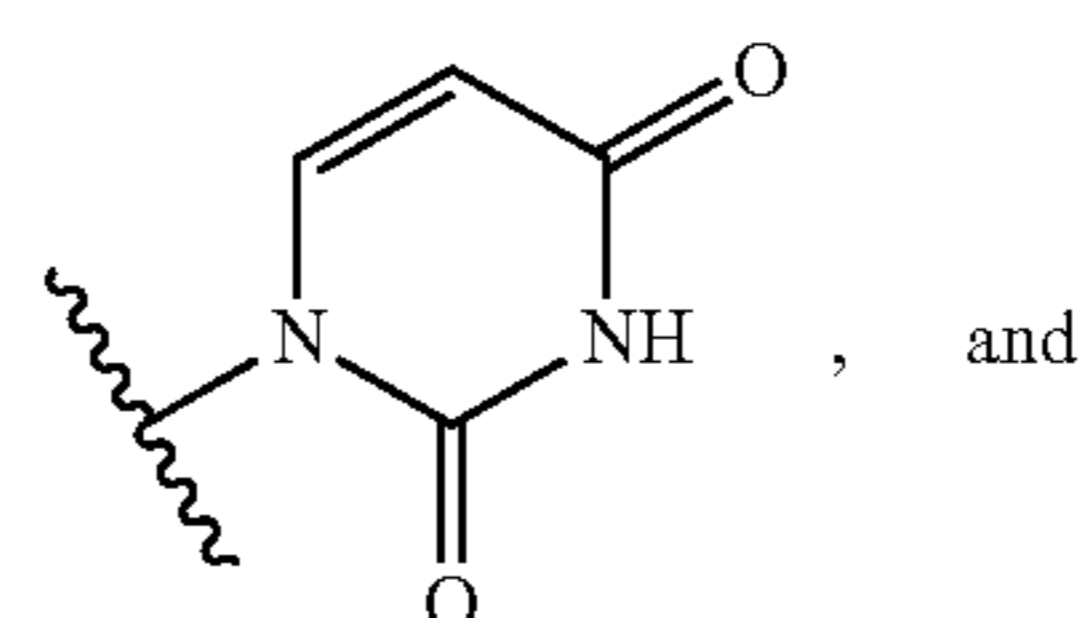
45 (A)



50 (5mC)



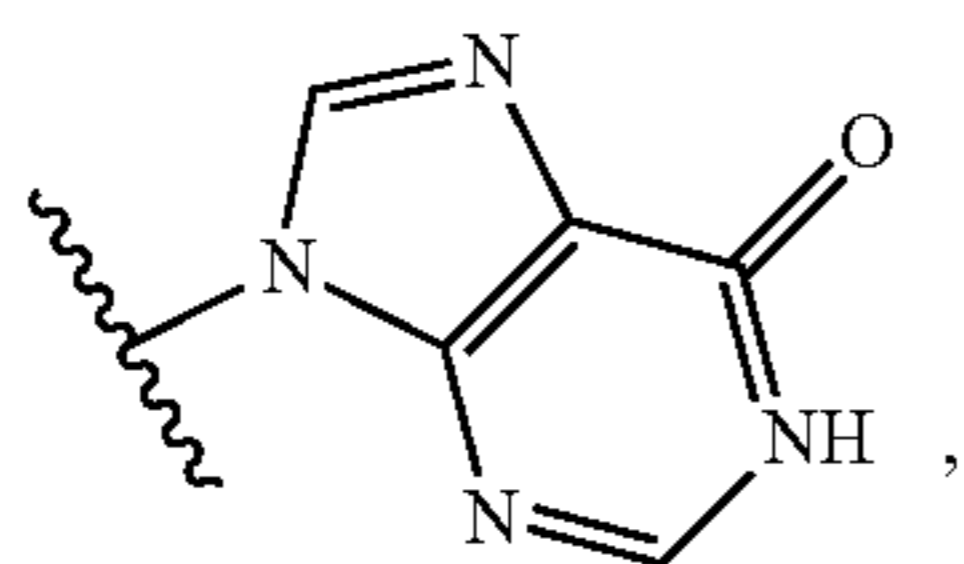
55 (U)



65

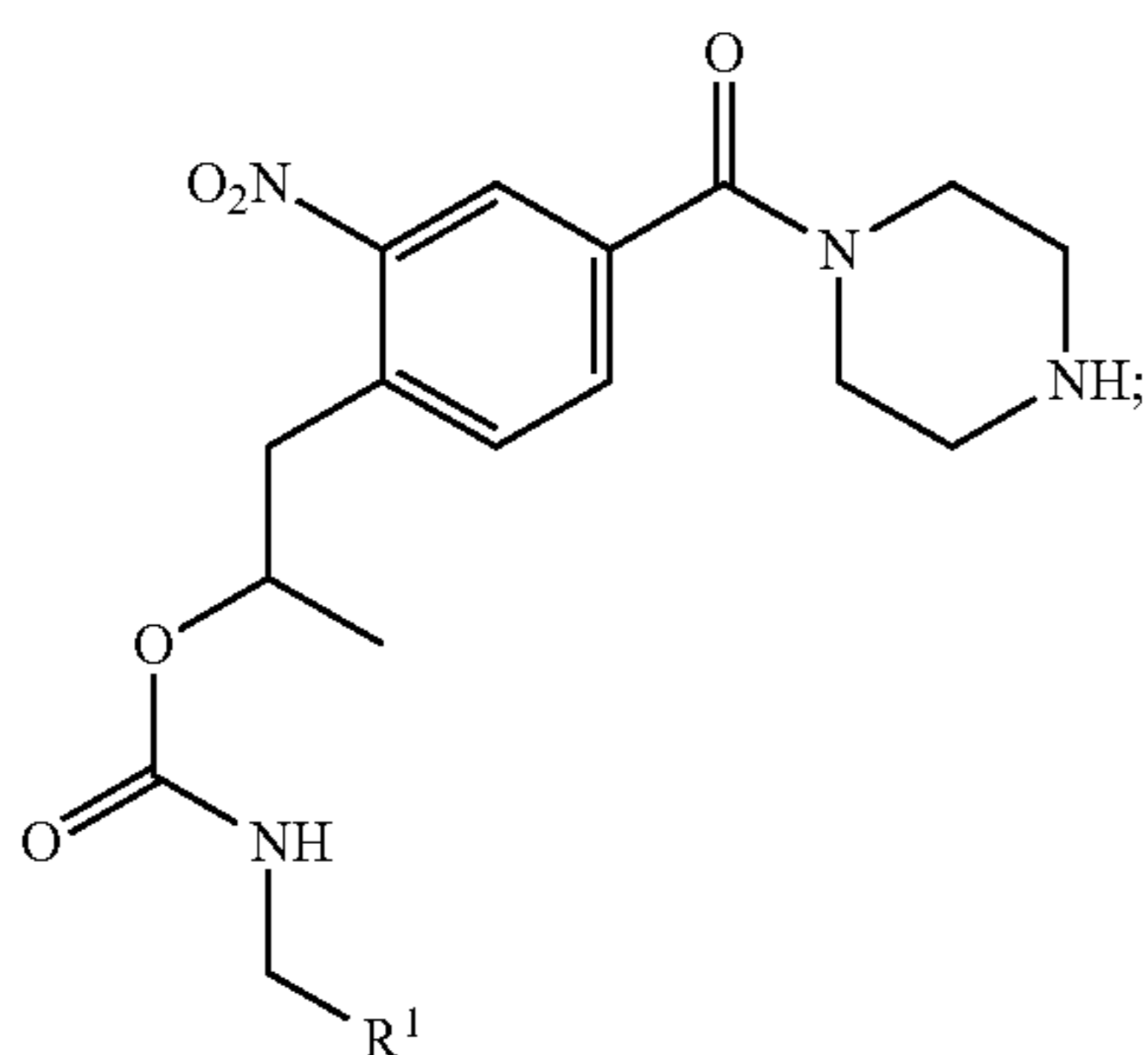
97

-continued

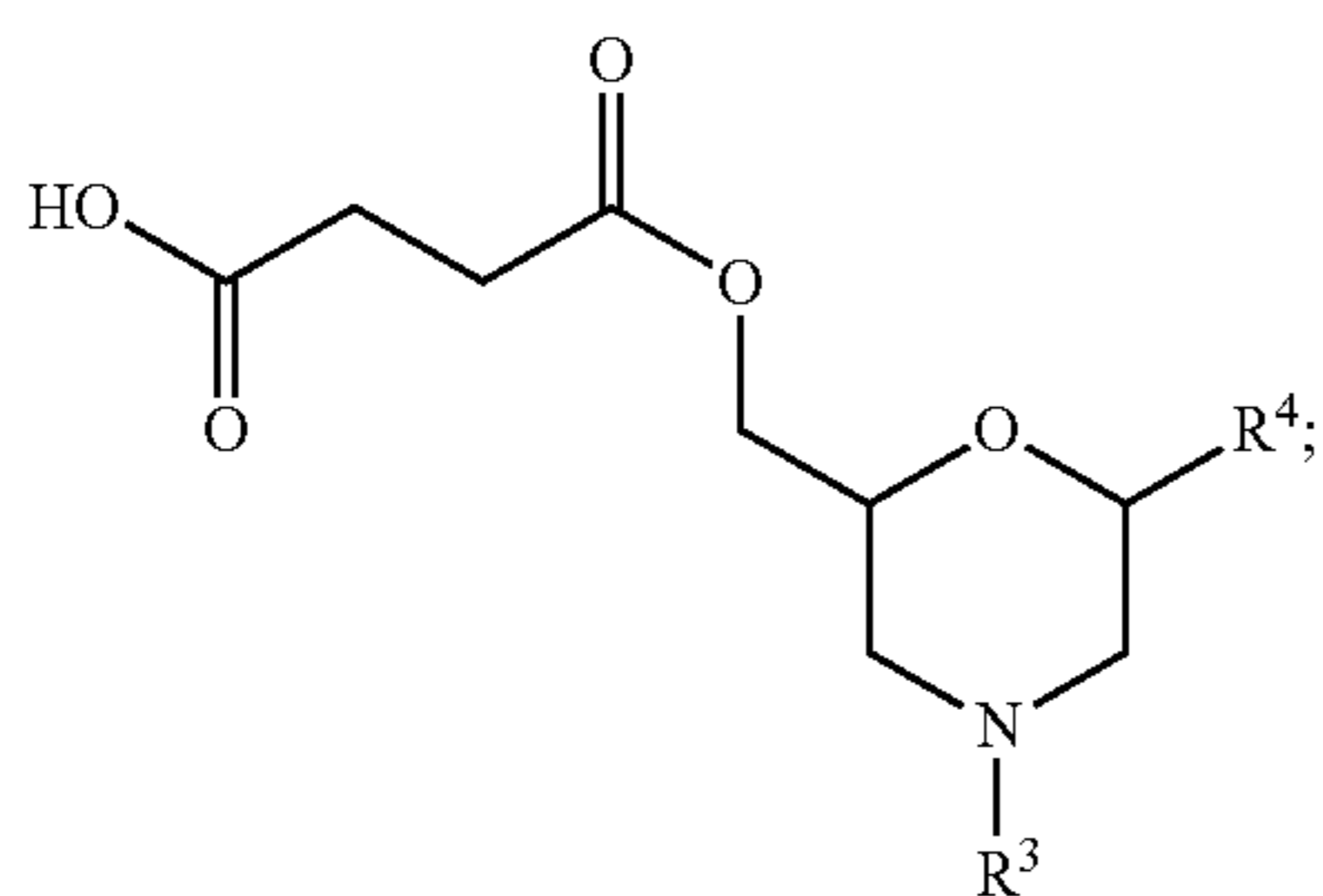


wherein the process comprises the sequential steps of:

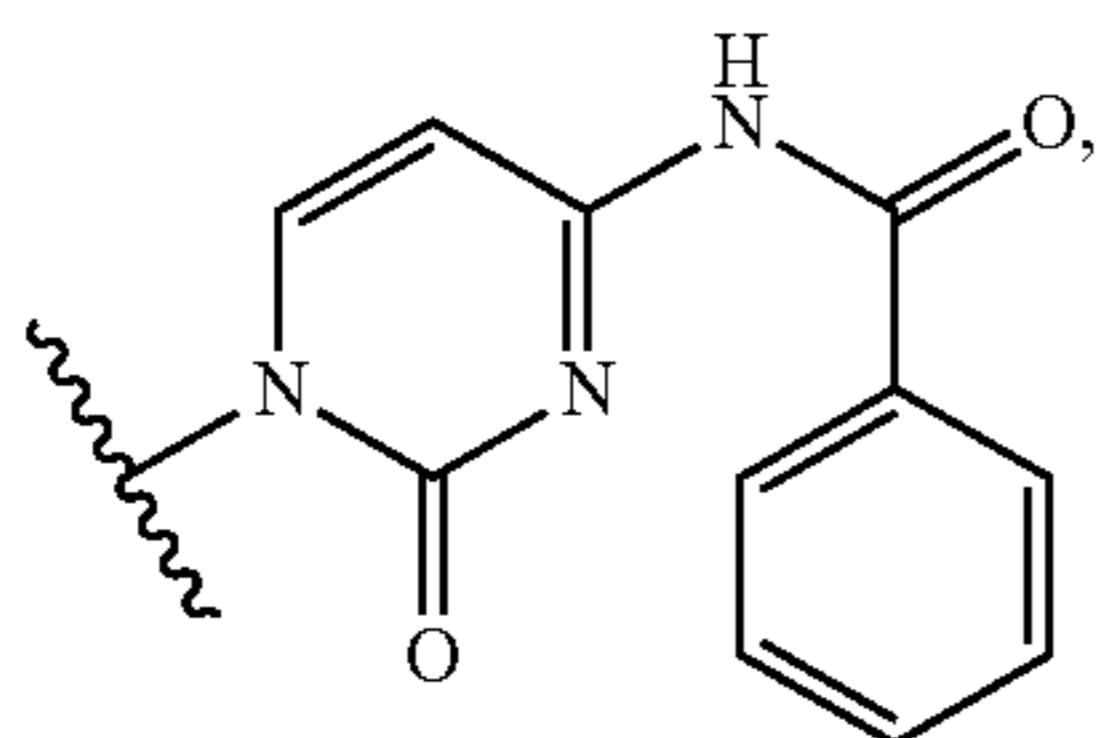
(a) contacting a compound of Formula (II):



wherein R¹ is a support-medium;
with a compound of Formula (S2):



wherein R⁴ is selected from the group consisting of:



98

-continued

(I)

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10

15

(II)

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40

(S2)

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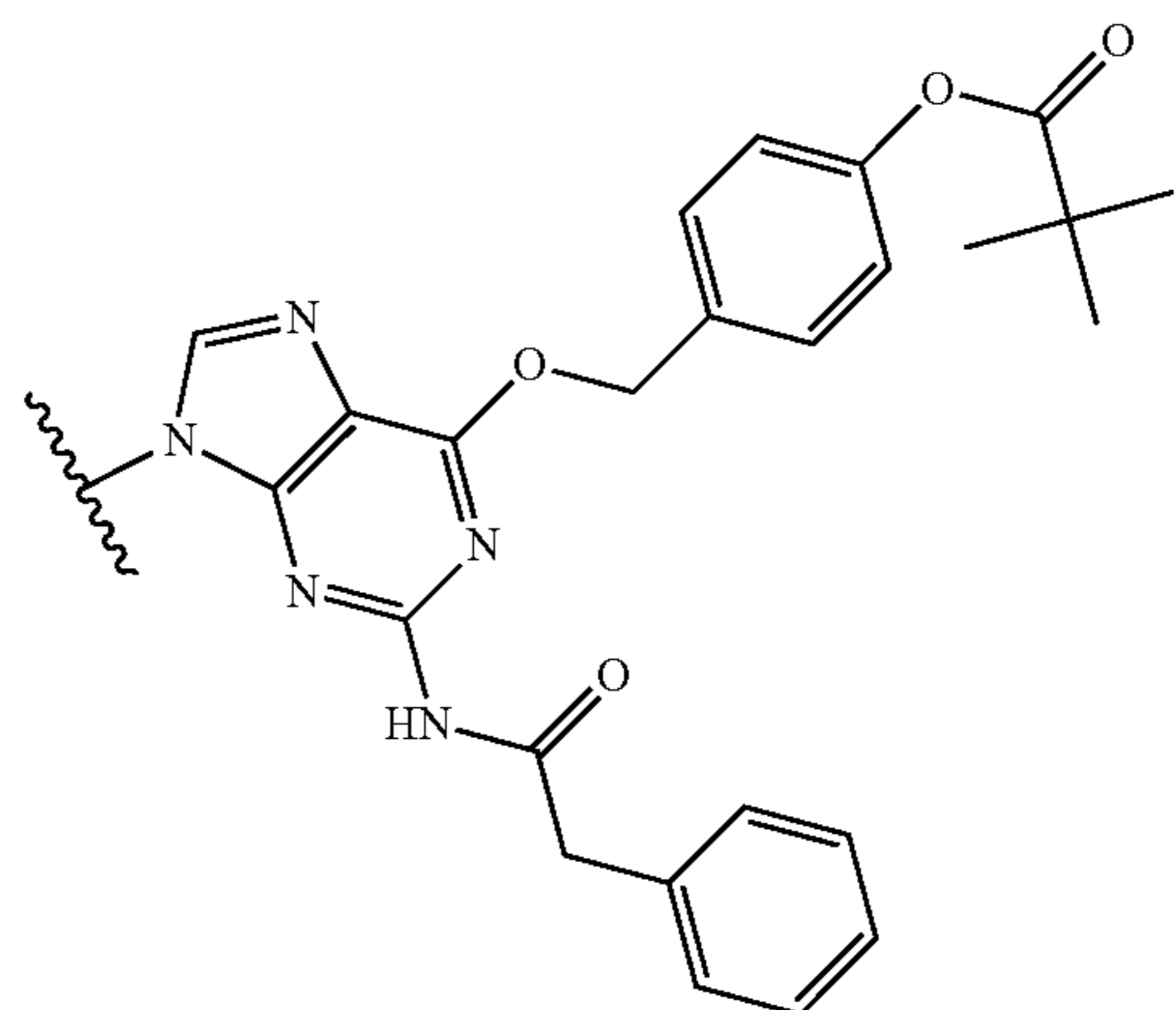
55

(PC)

60

65

(DPG)



(T)

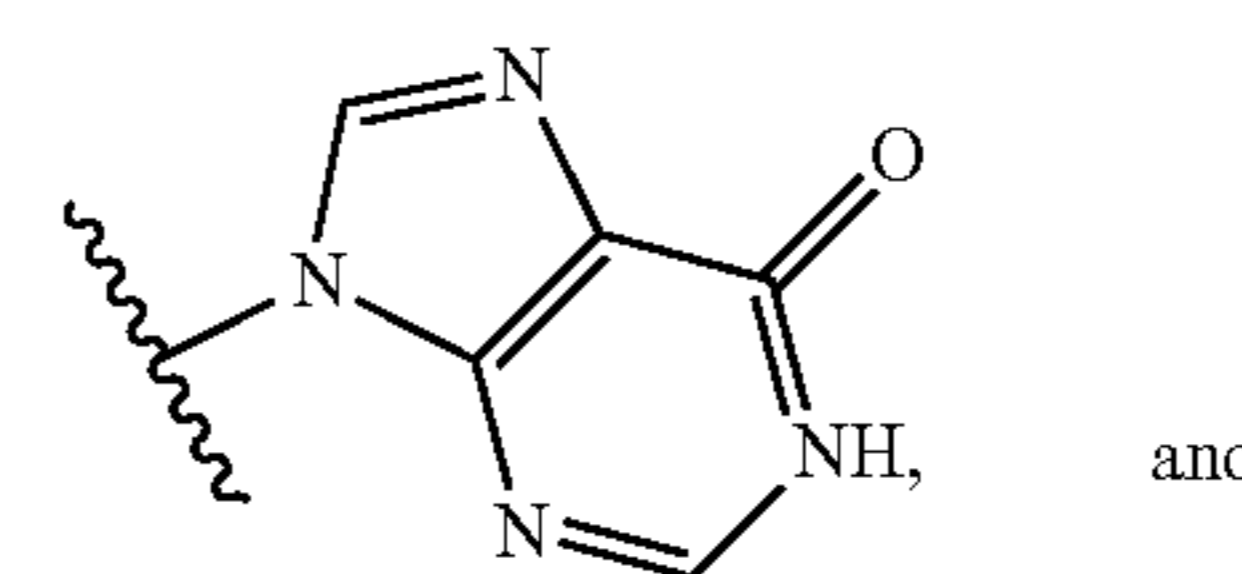
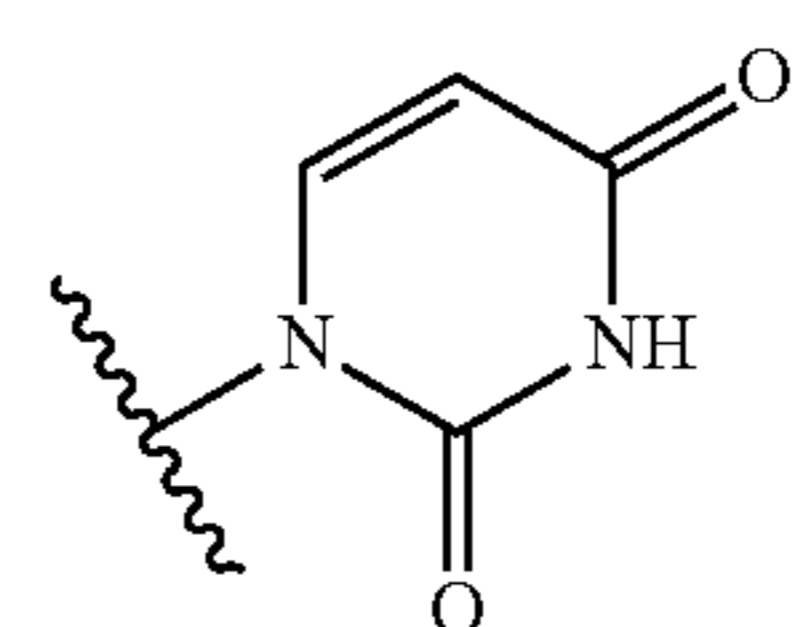
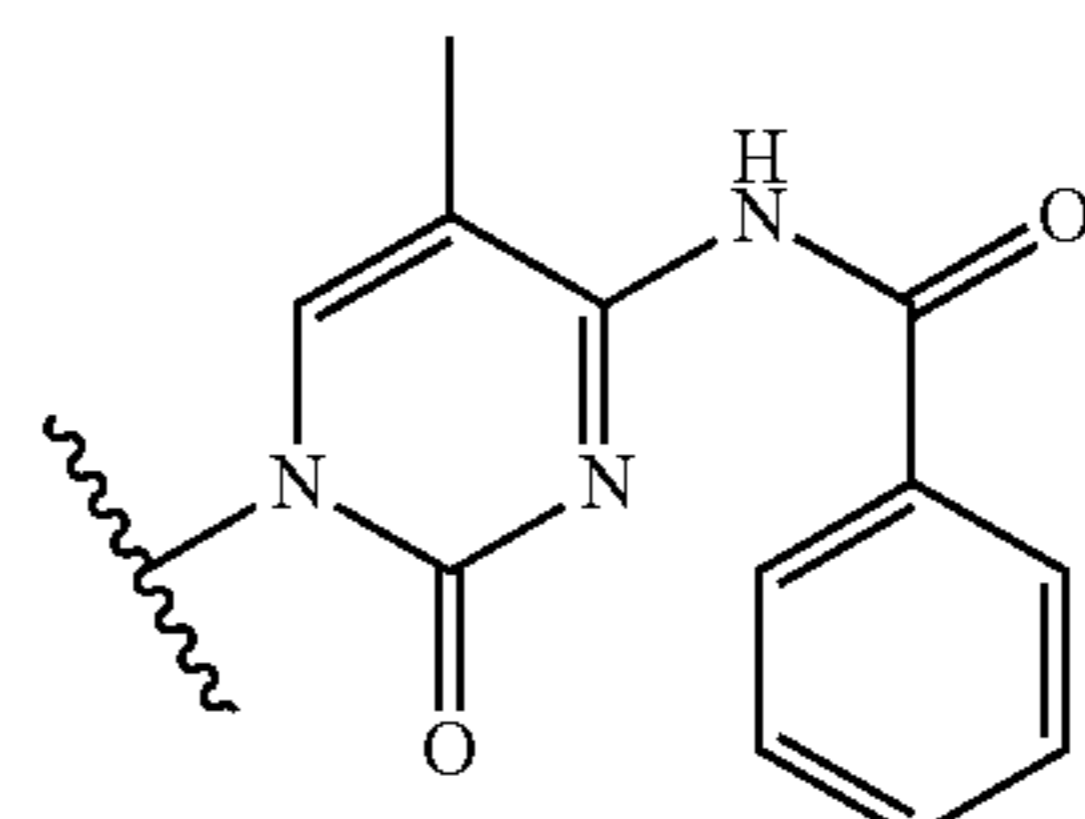
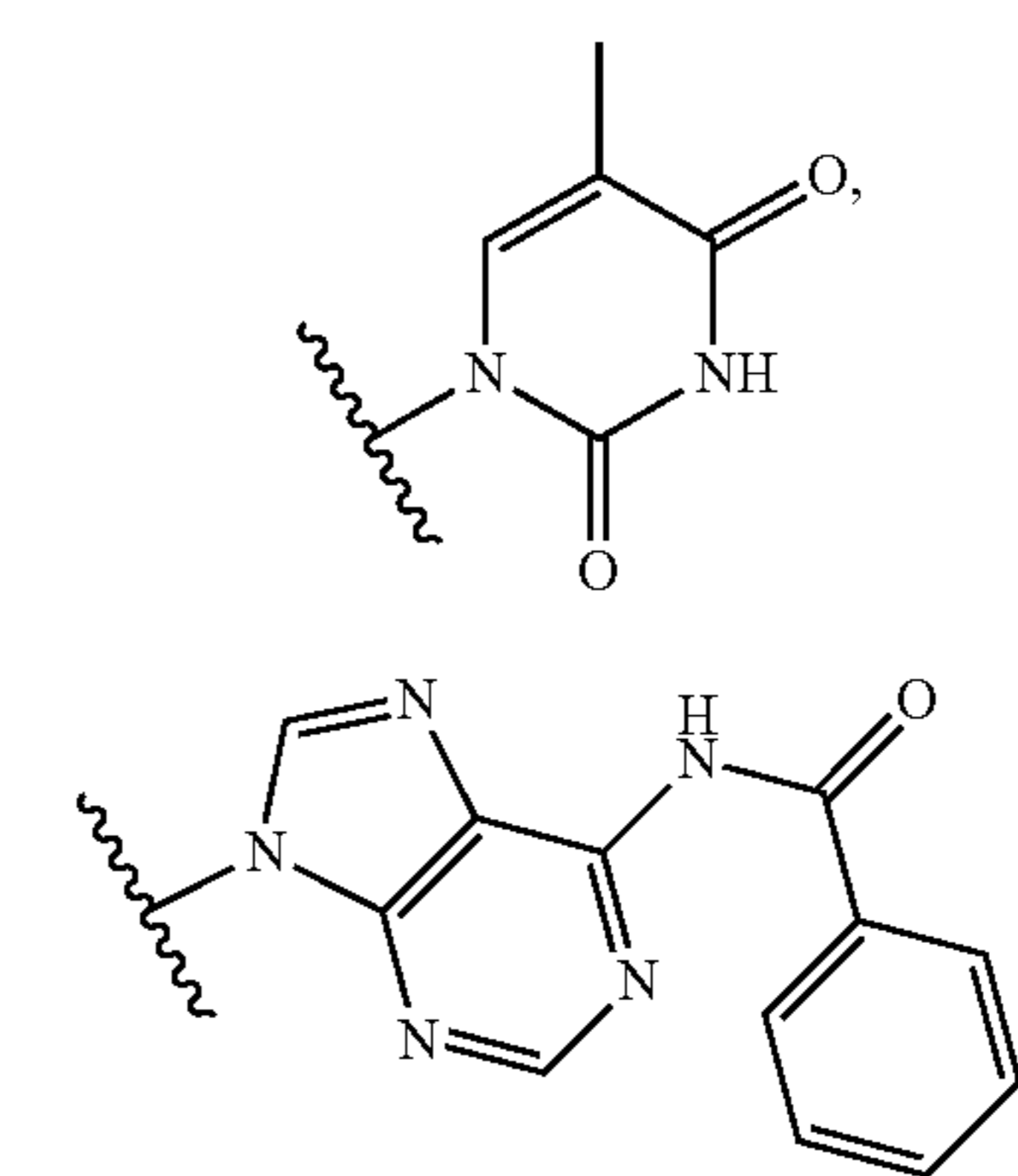
(PA)

(P5mC)

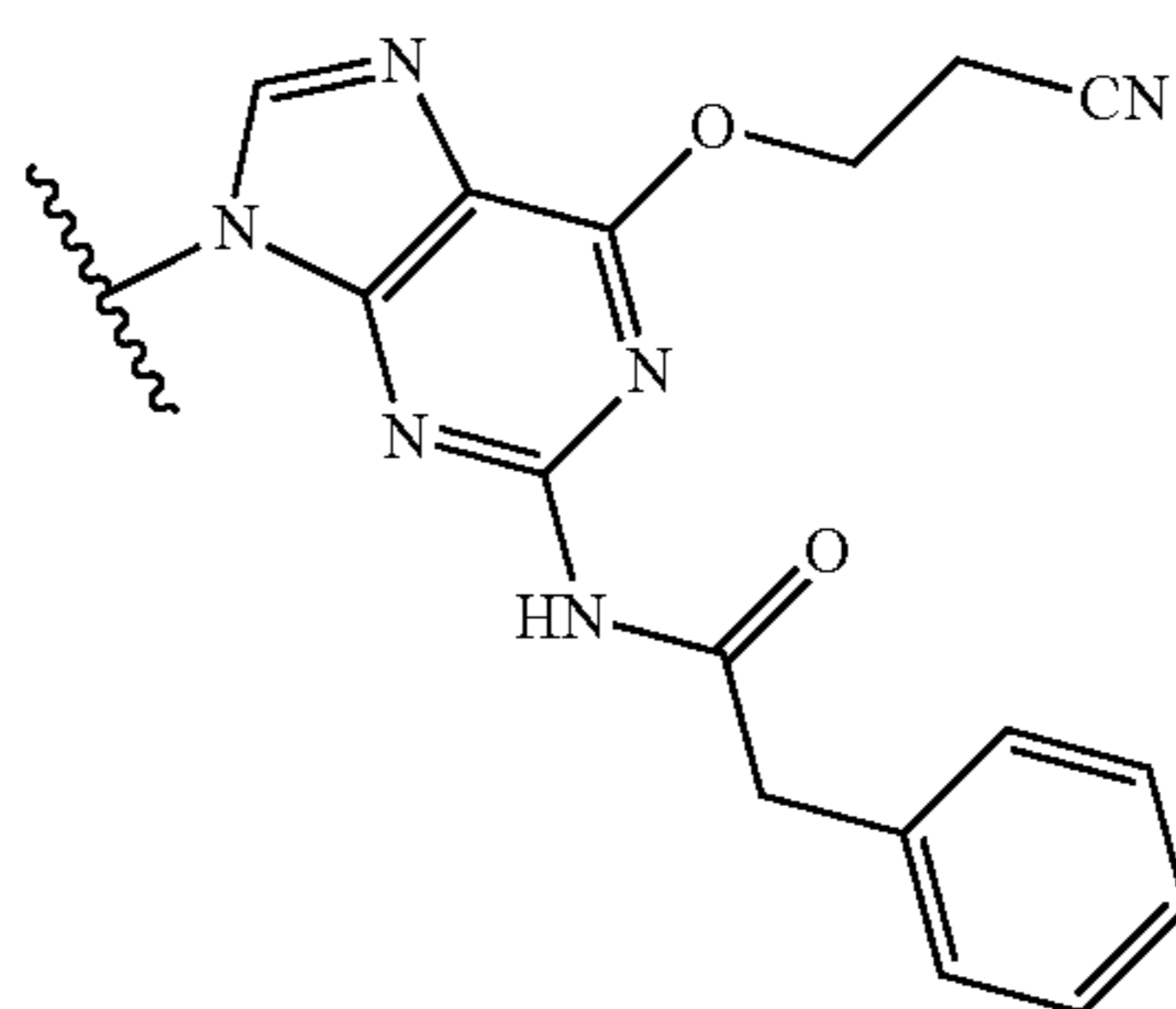
(U)

(I)

(PG)



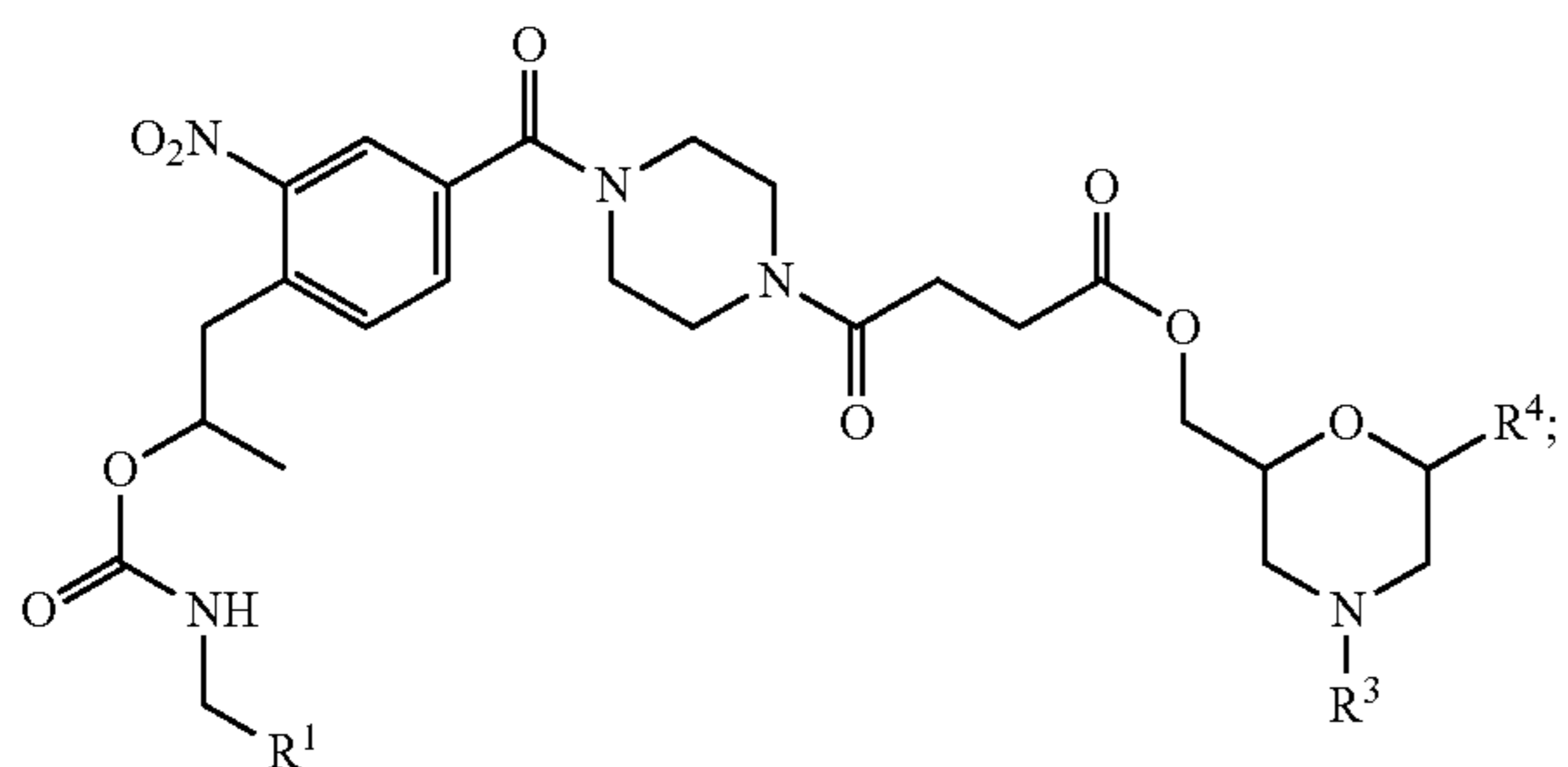
and



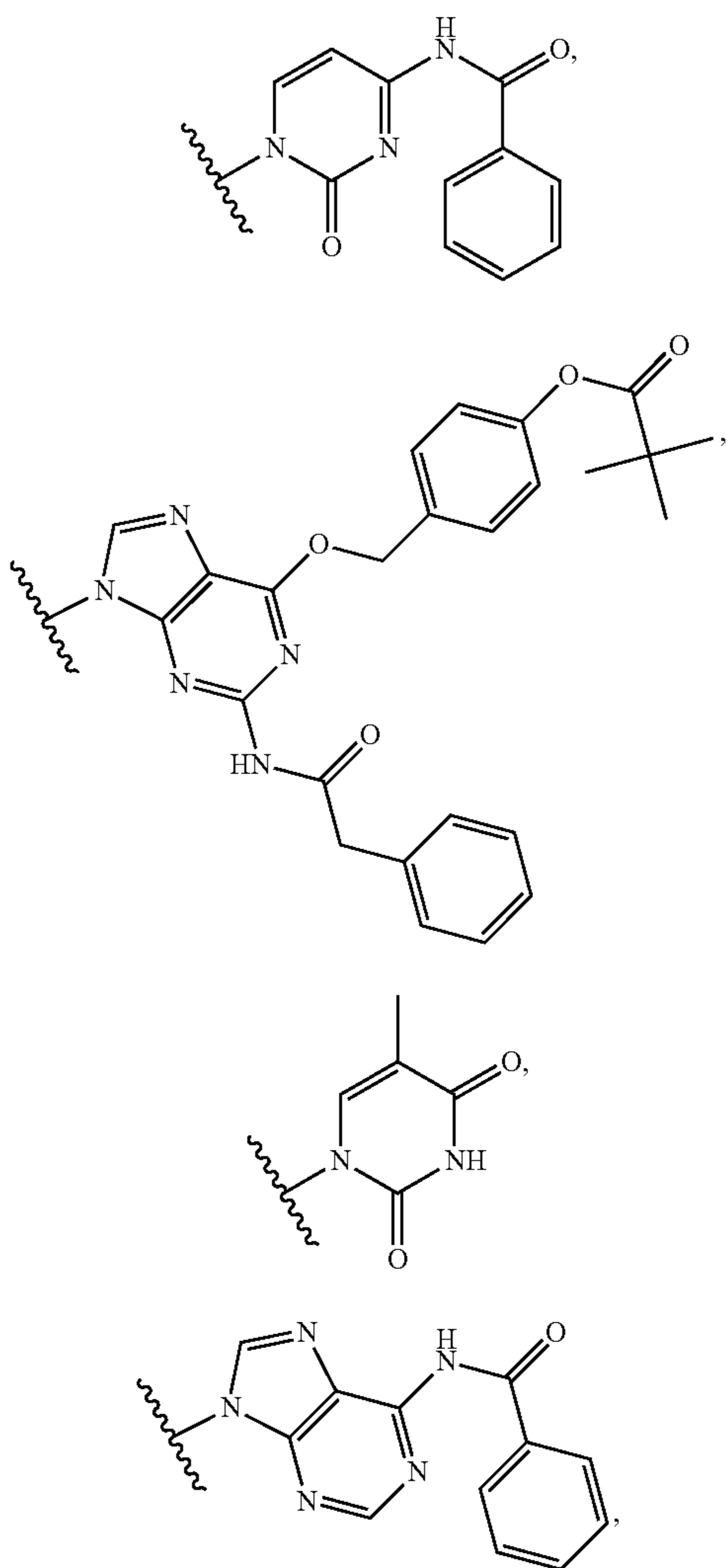
and

99

wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
to form a compound of Formula (S3):



wherein R¹ is a support-medium, R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

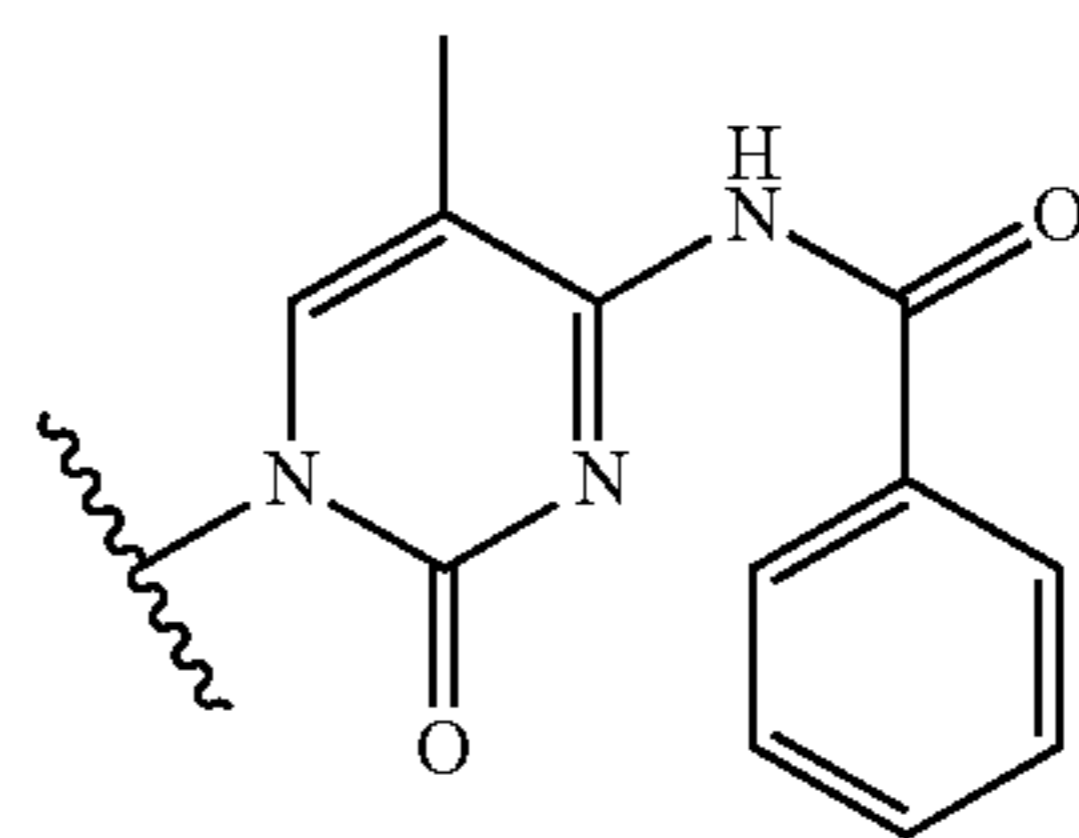


100

-continued

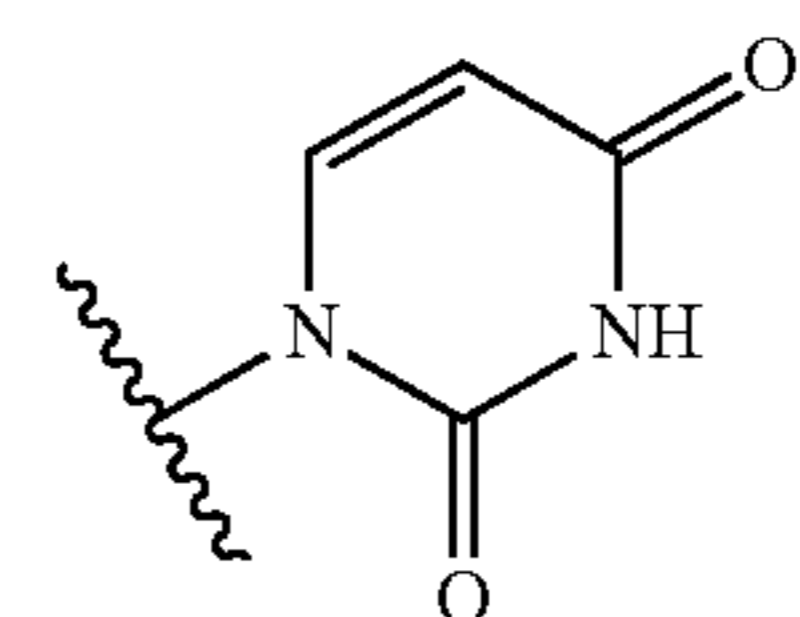
(P5mC)

5



(S3)

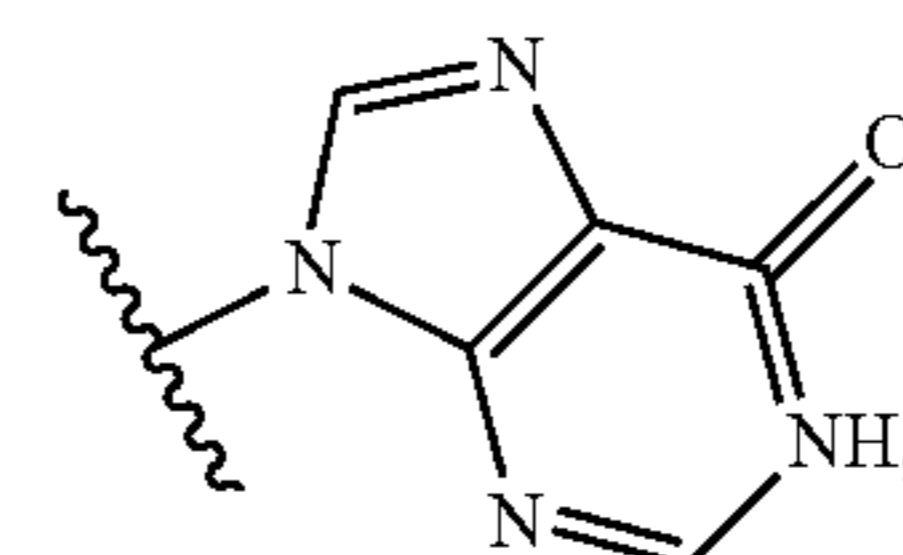
10



(U)

15

20

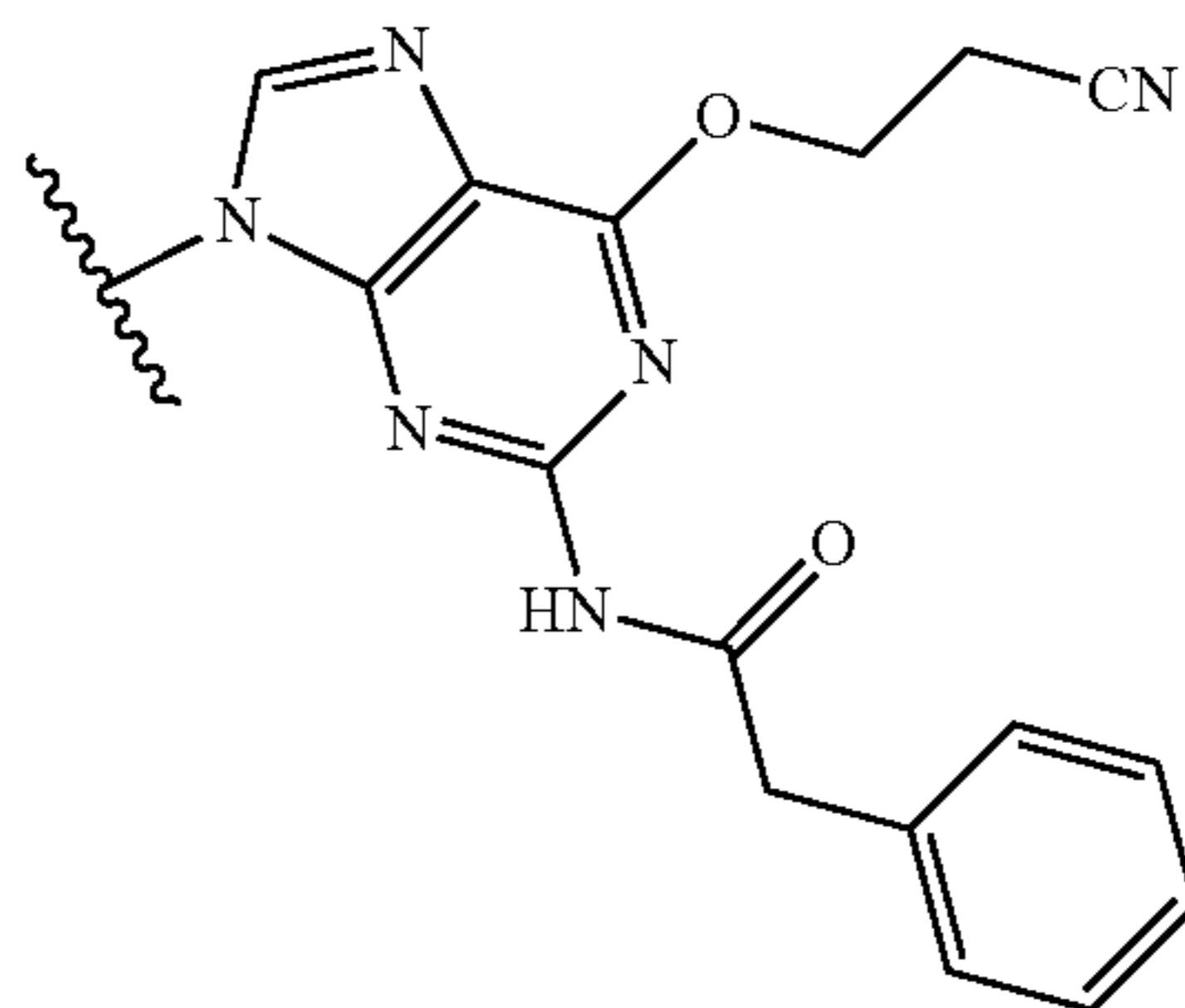


(I)

and

(PG)

25



(PC)

30

35

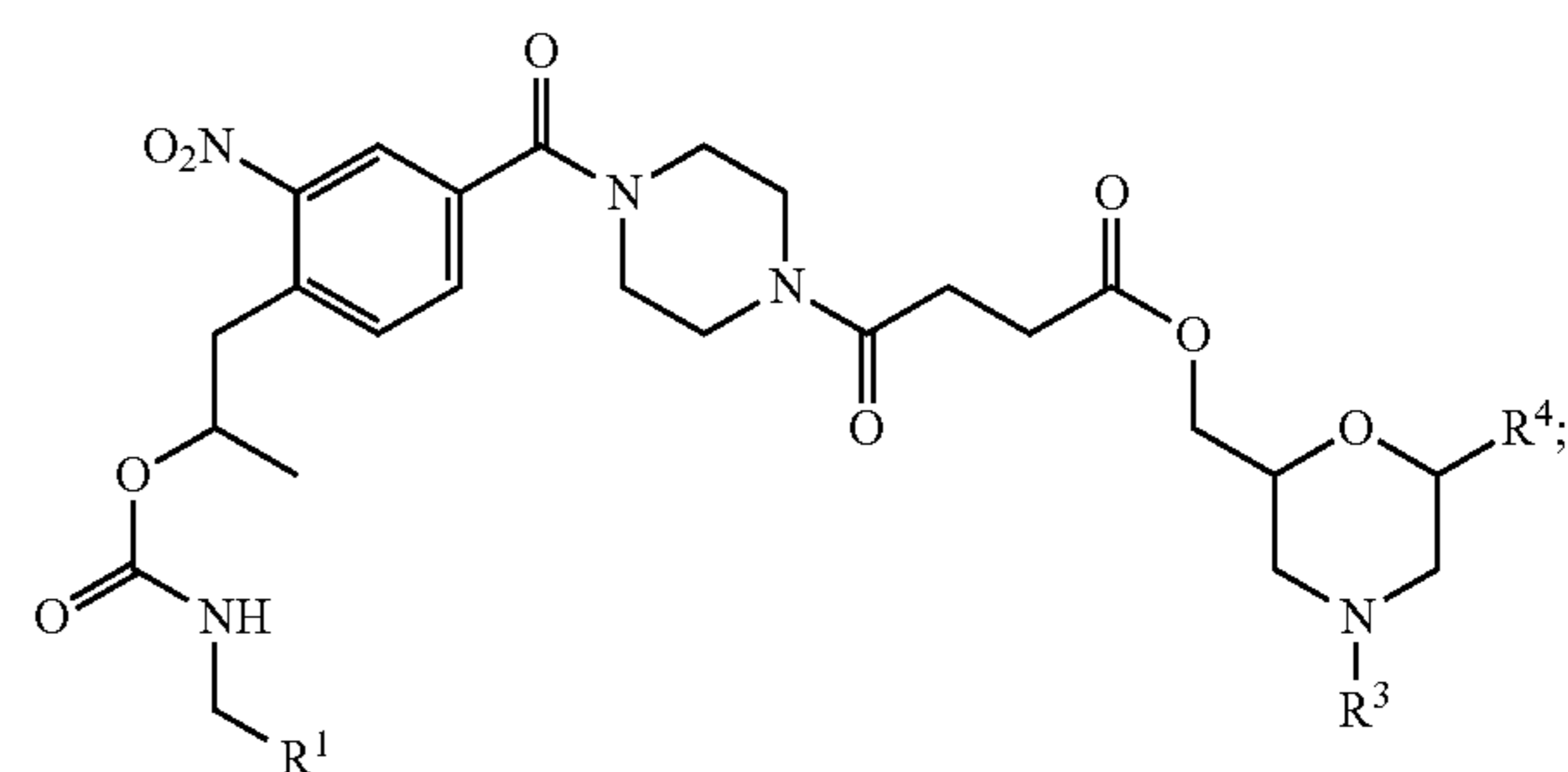
(b) contacting the compound of Formula (S3) with a deblocking agent to form a compound of Formula (IV-S):

(DPG)

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(IV-S)

45



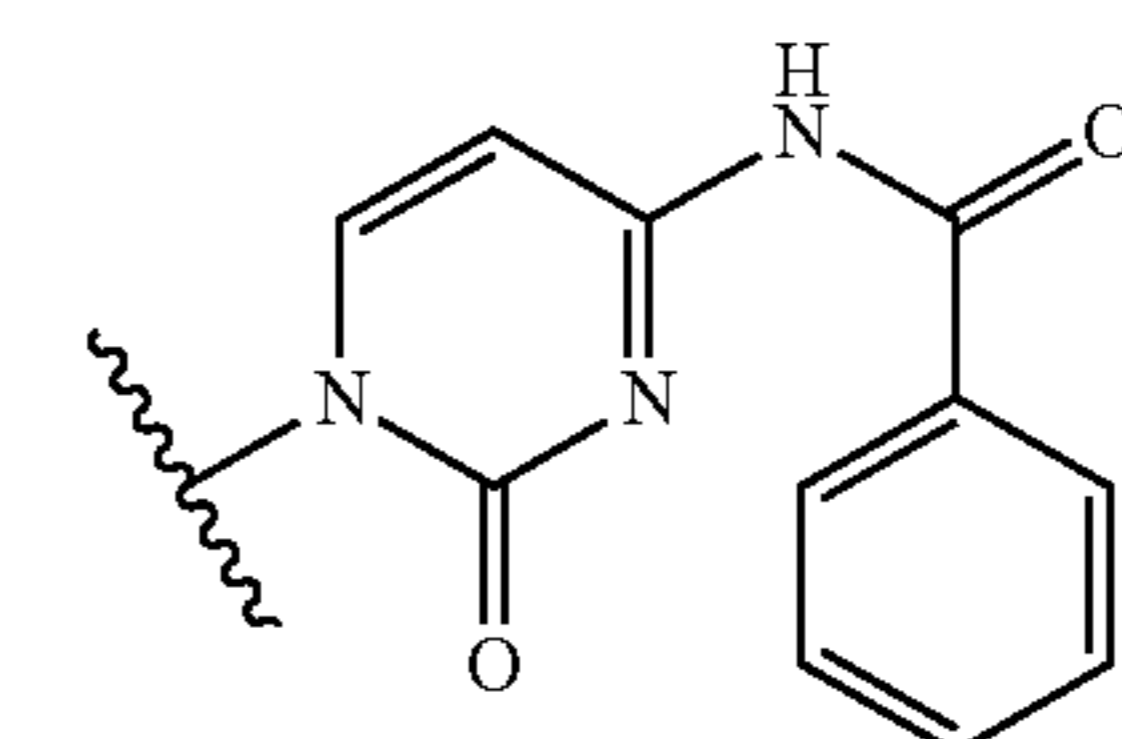
(T)

55

wherein R¹ is a support-medium, R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

(PA)

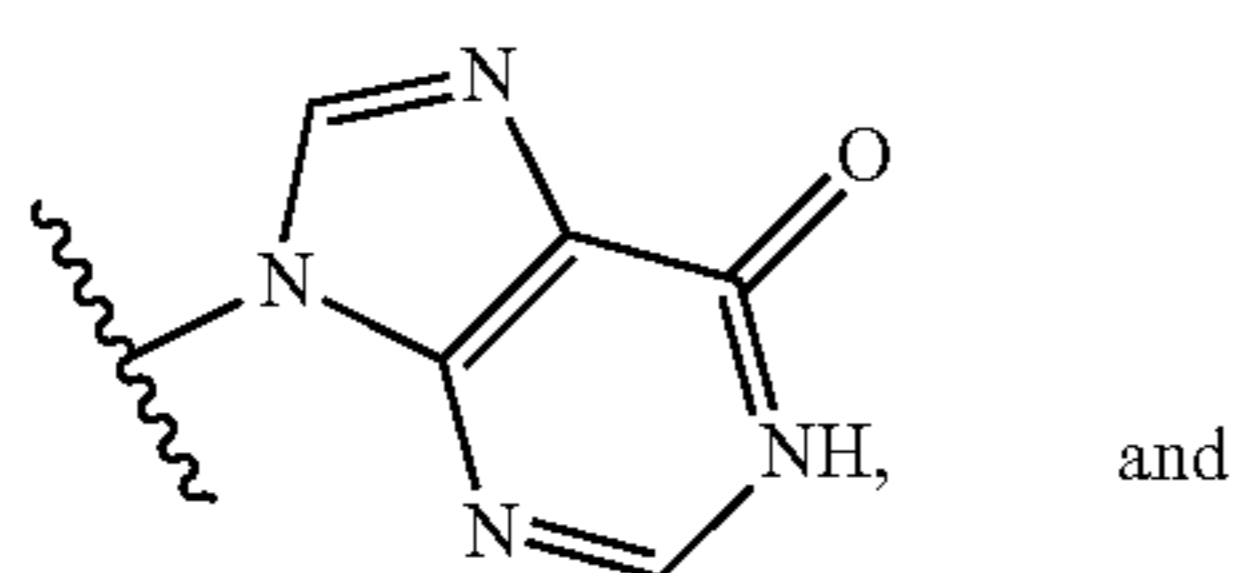
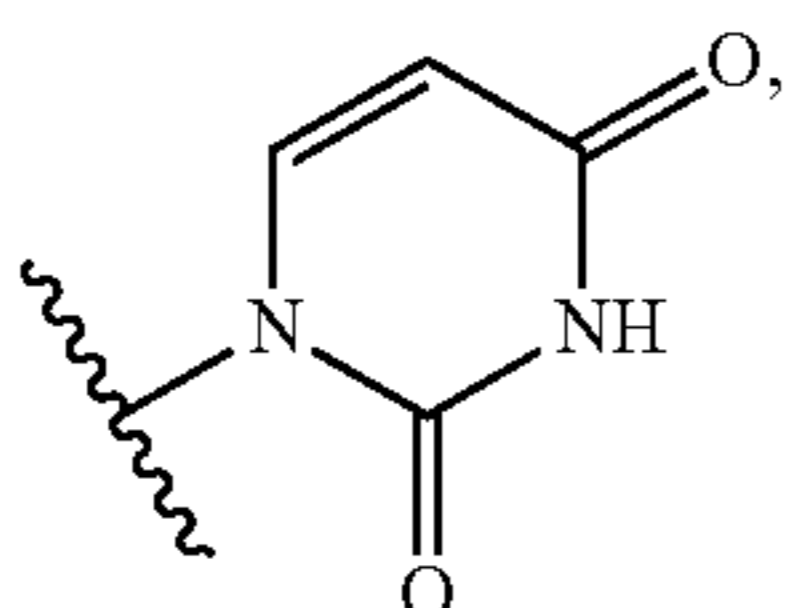
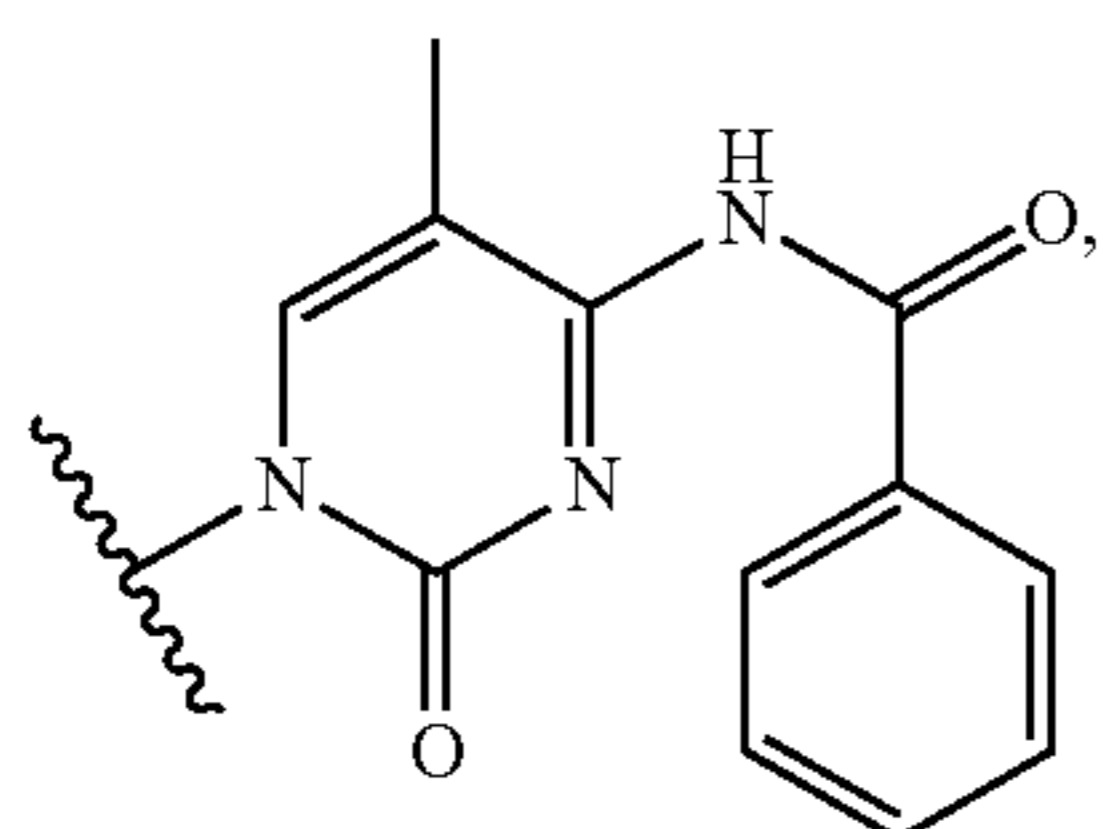
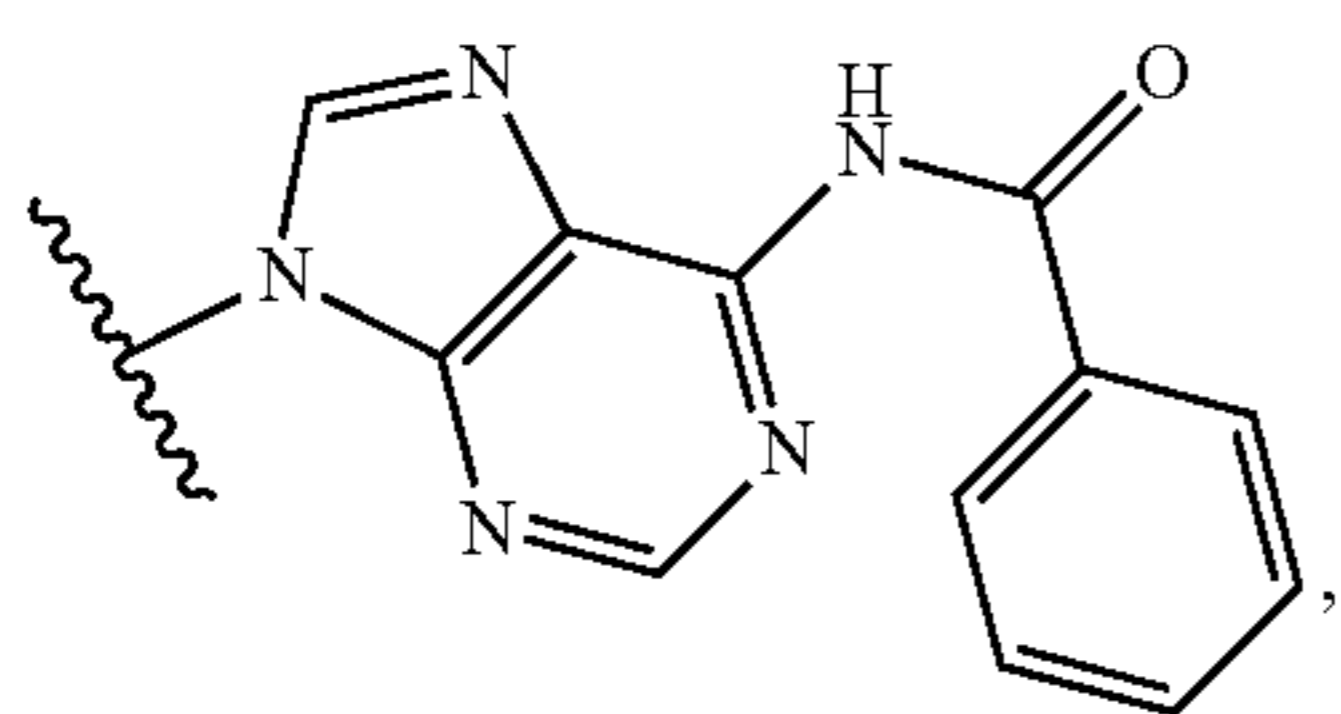
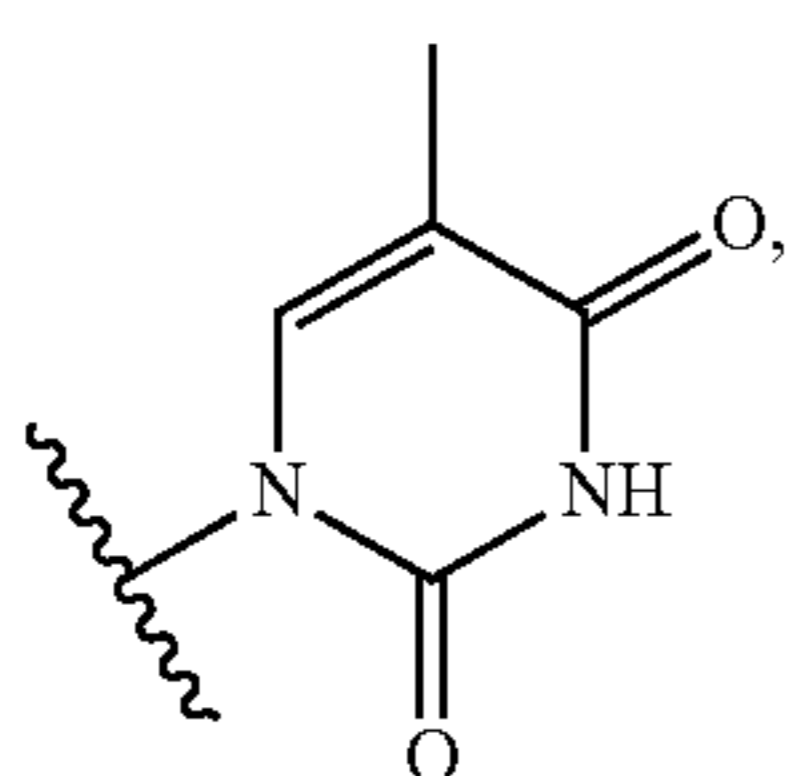
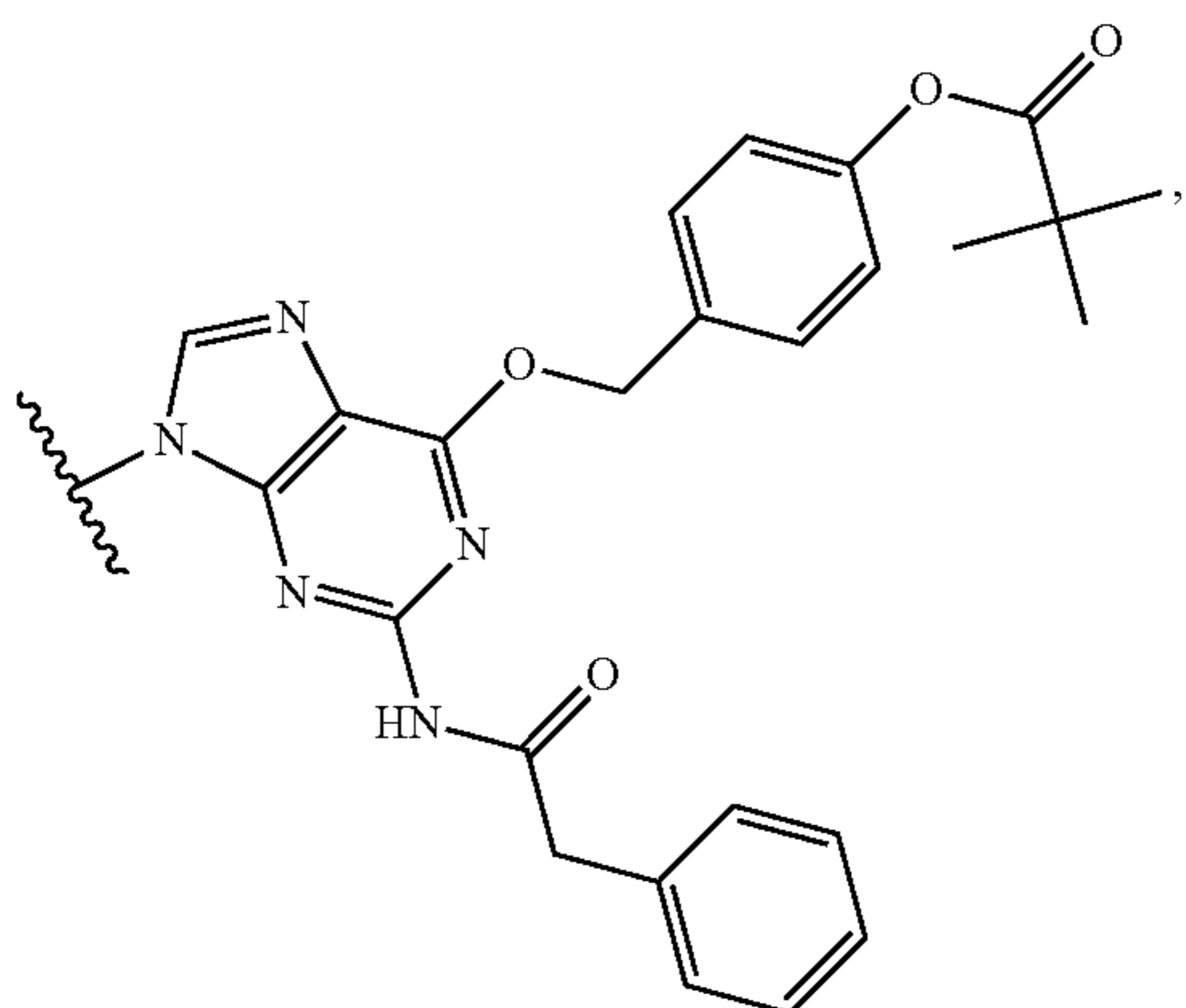
65



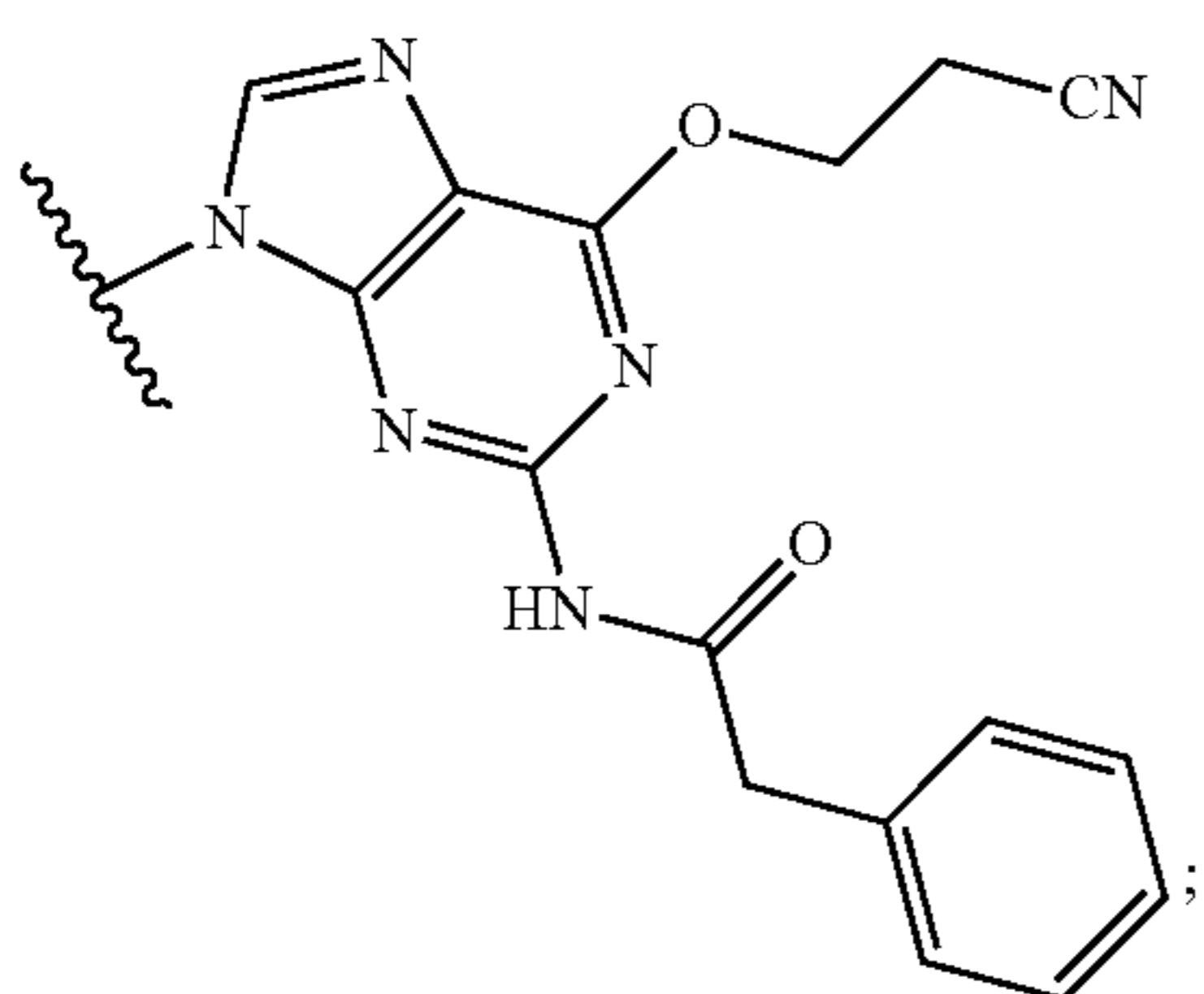
(PC)

101

-continued



and



102

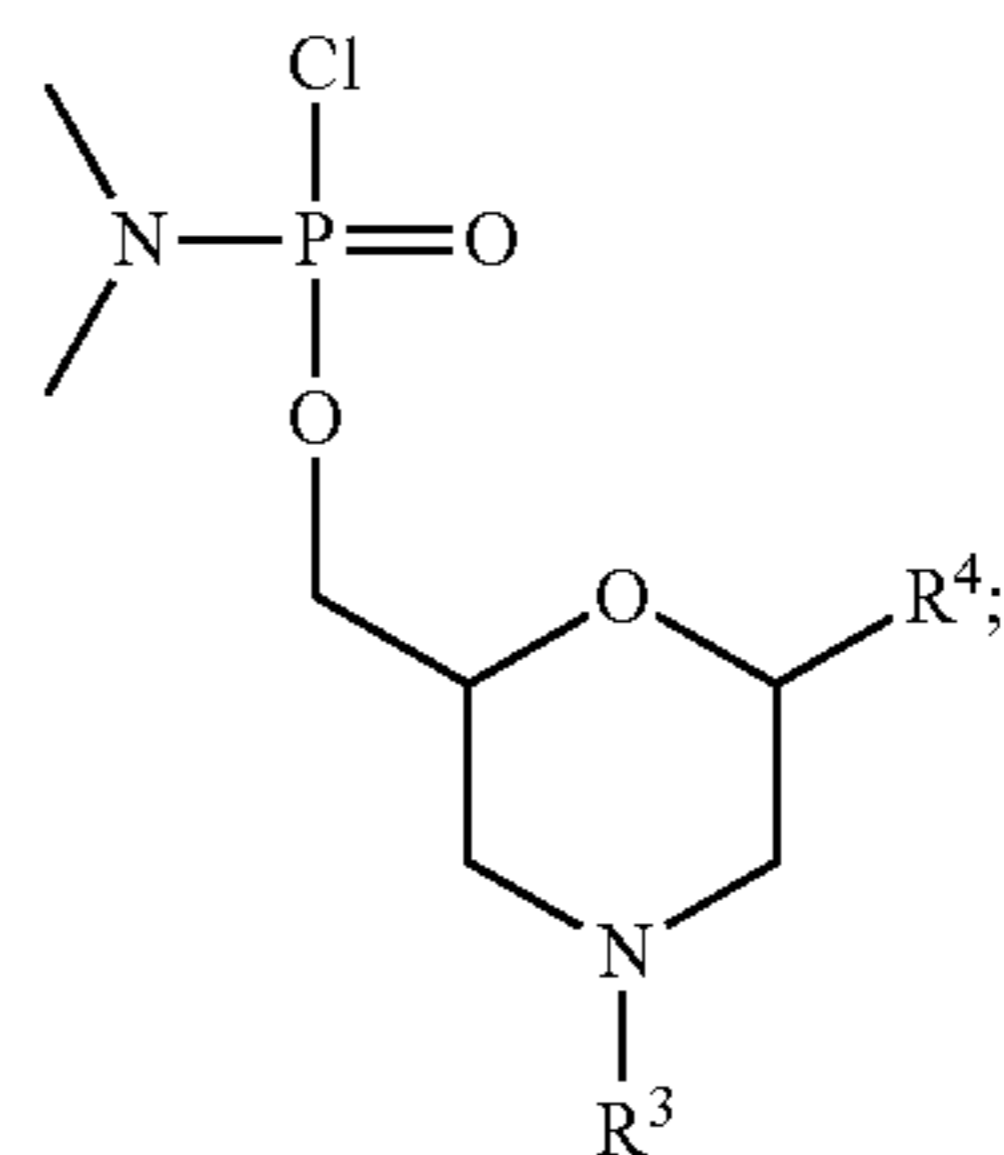
(c) contacting the compound of Formula (IV-S) with a compound of Formula (A4):

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(A4)

10

15



(T)

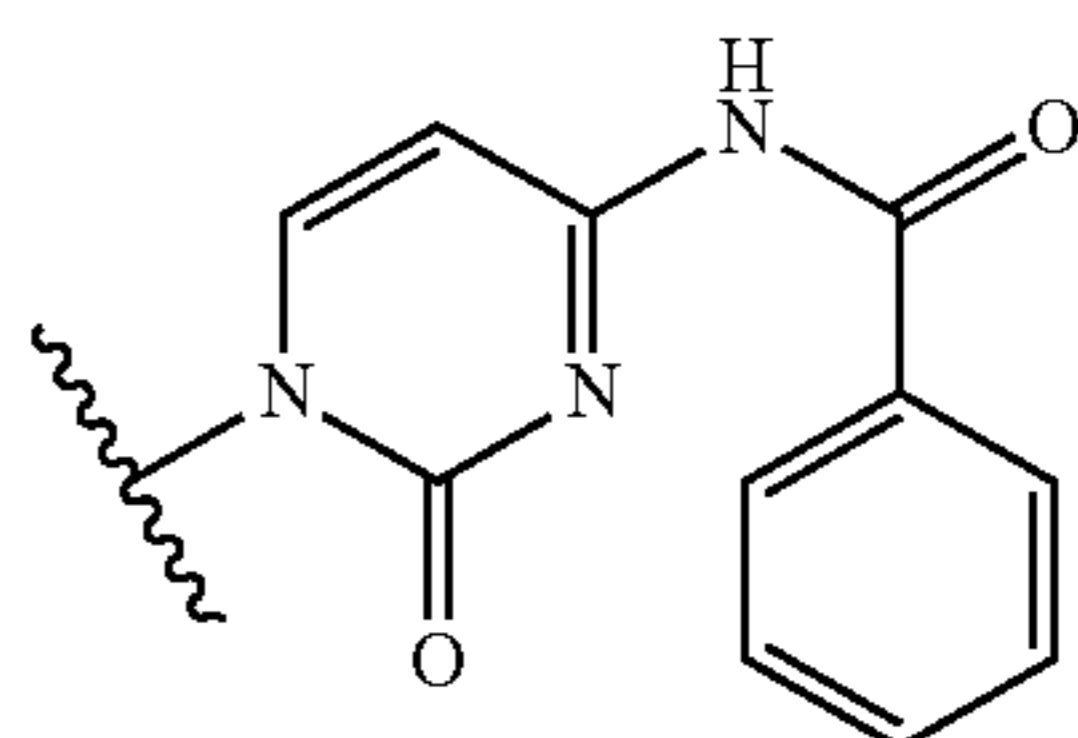
wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is selected from the group consisting of:

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(PC)

25

(PA)

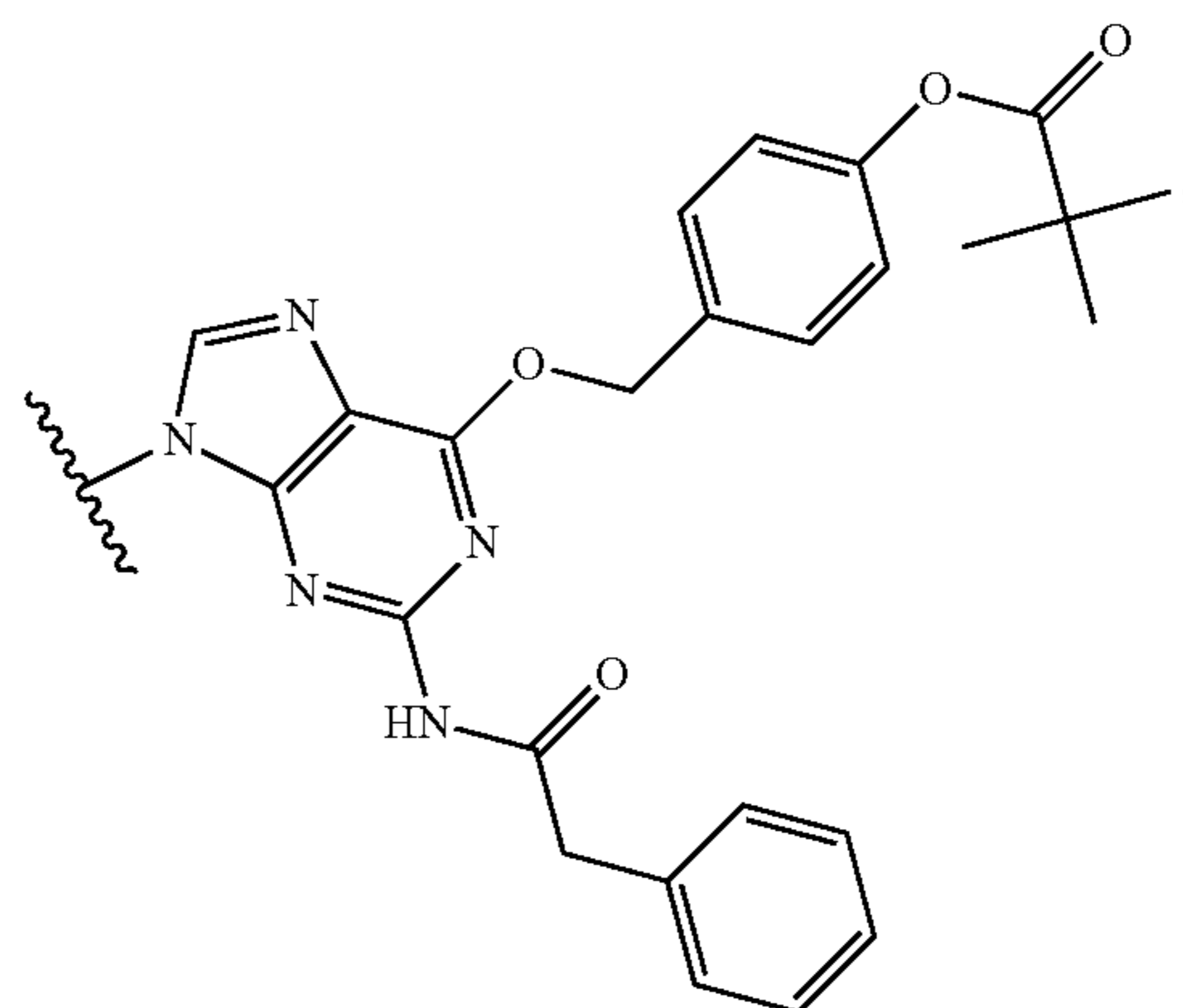


30

(DPG)

(P5mC)

35



40

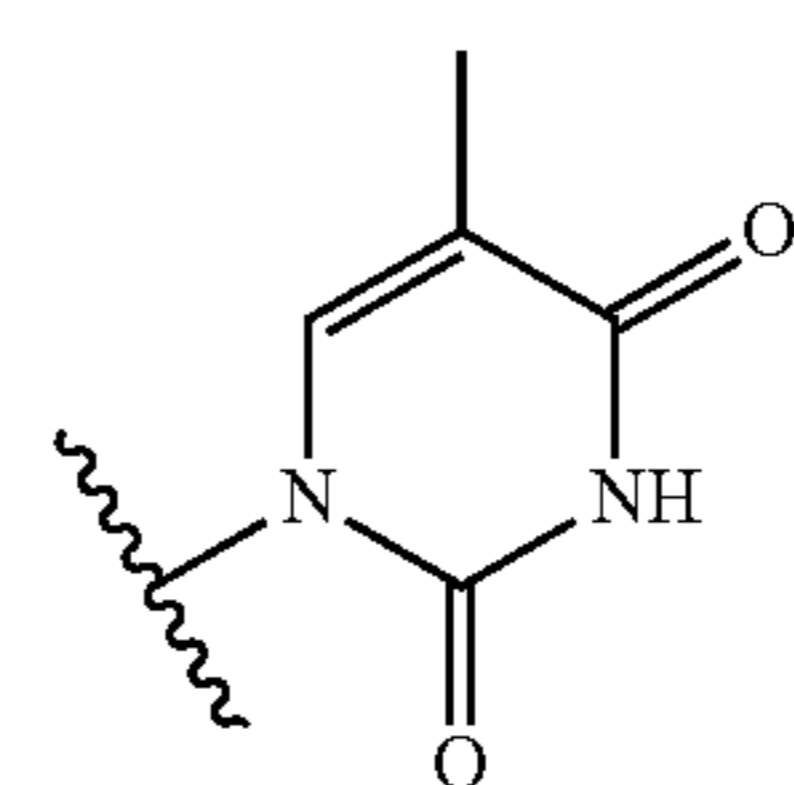
(U)

45

(I)

50

(T)

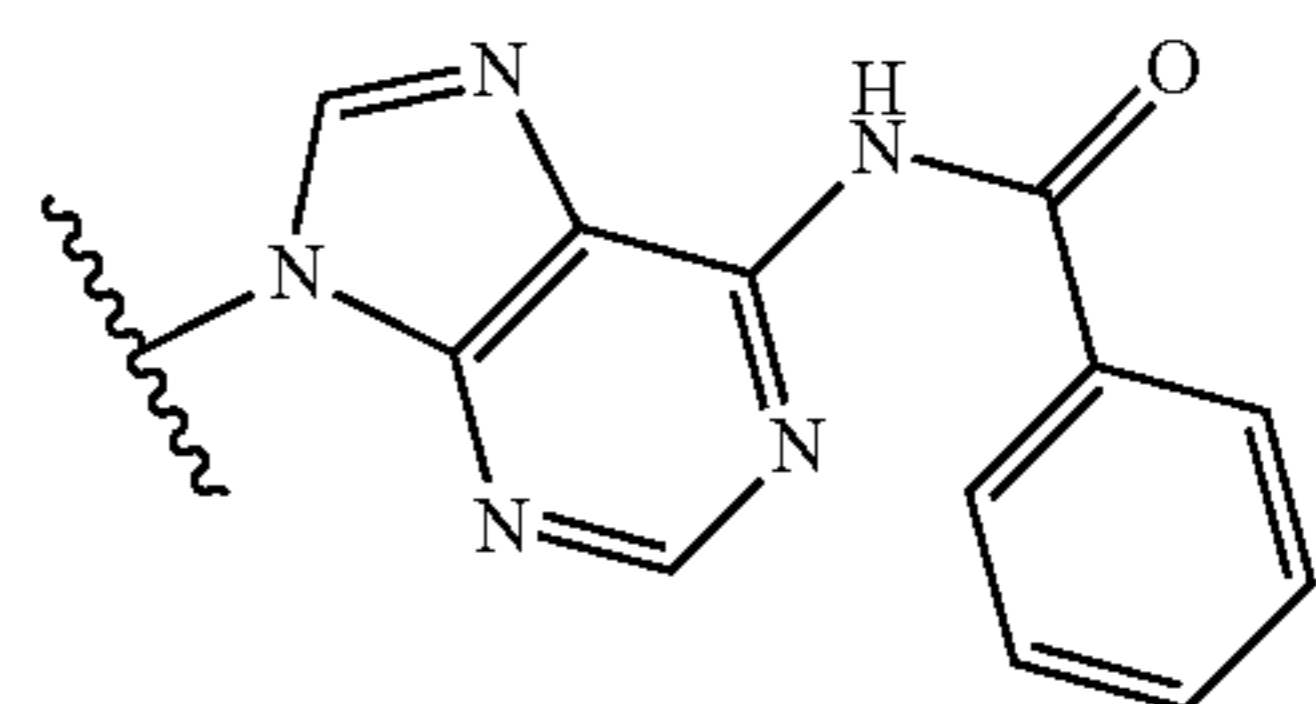


(PG)

55

(PA)

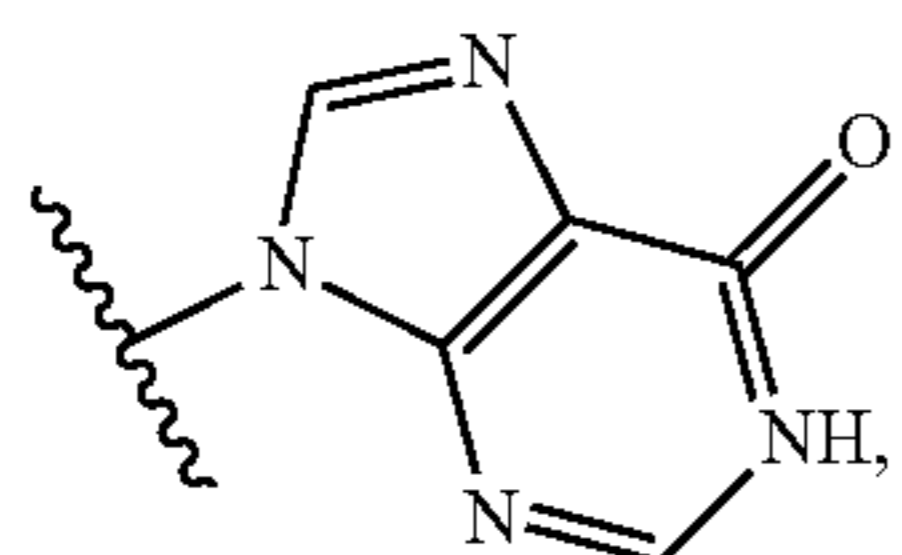
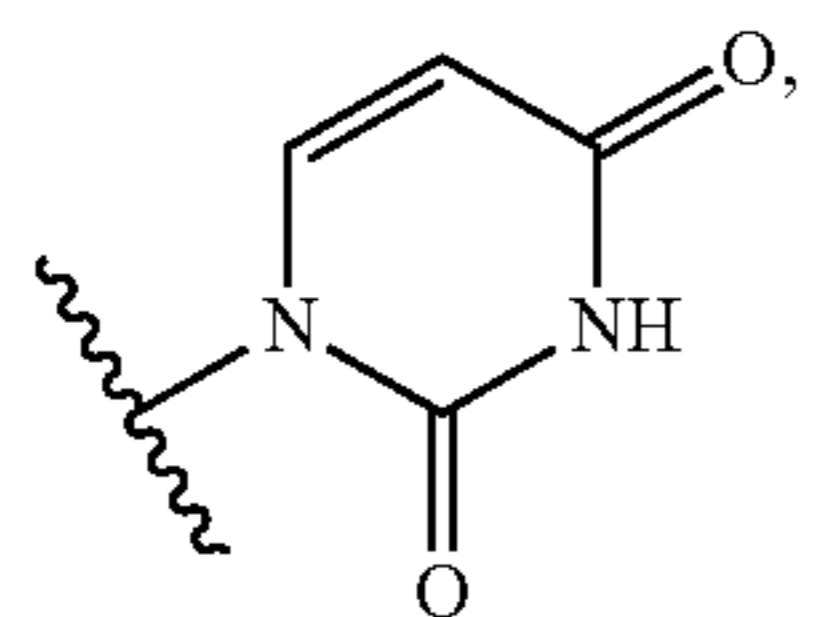
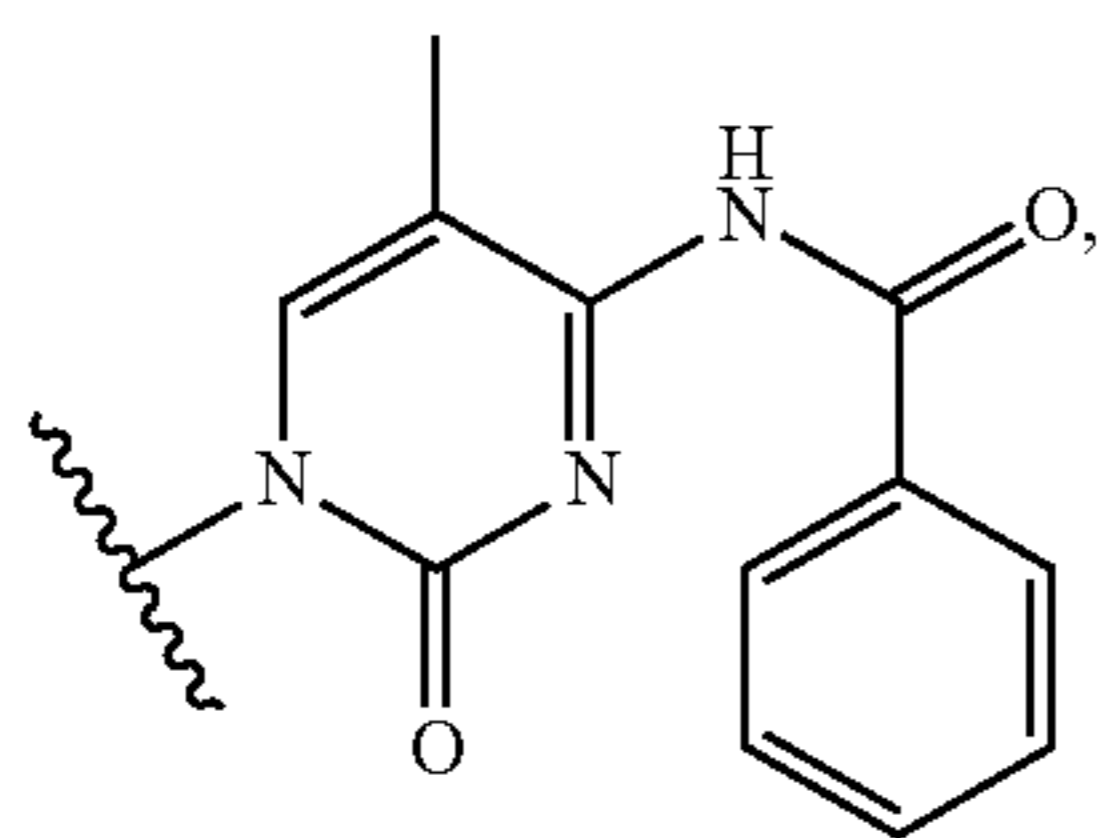
60



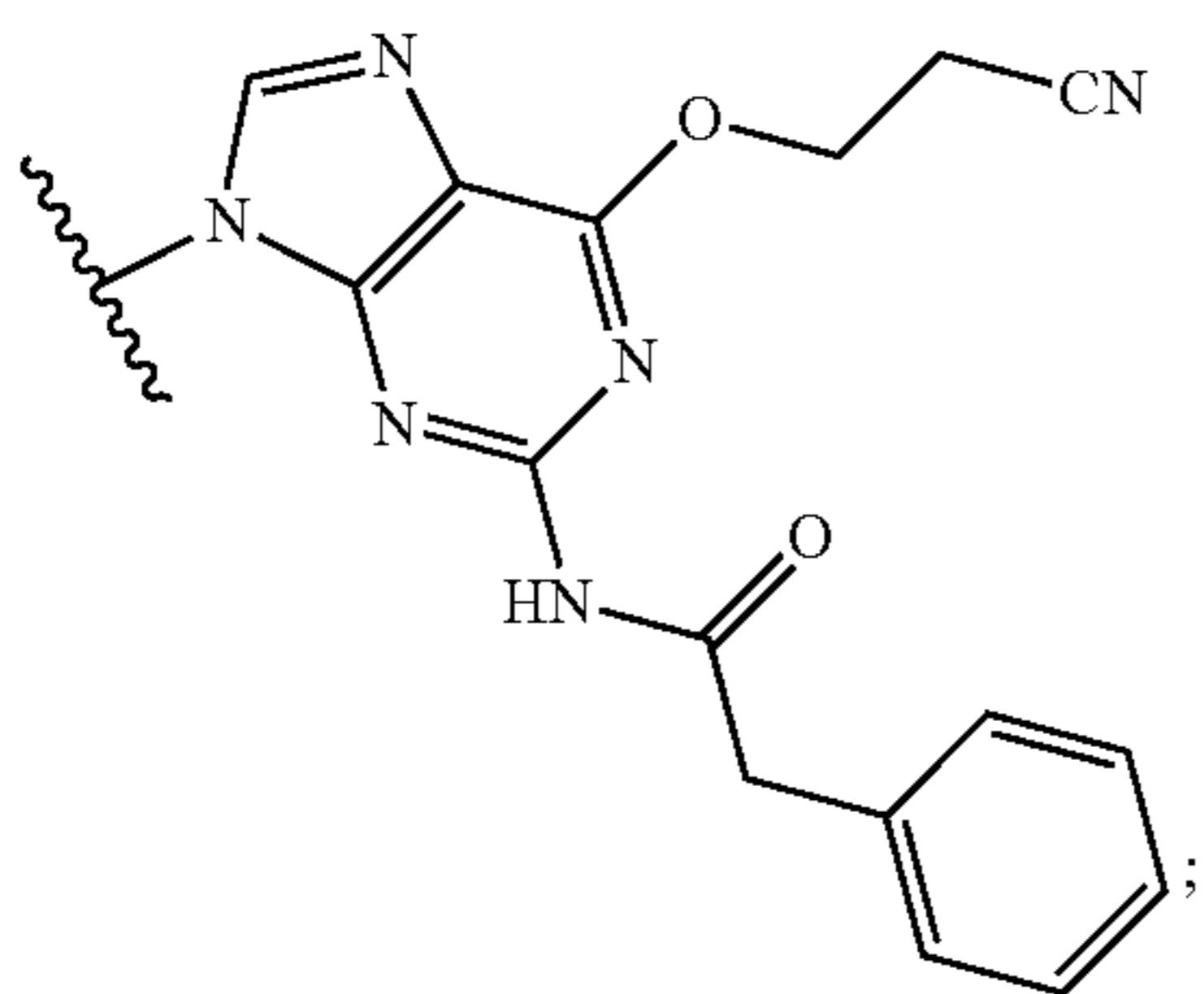
65

103

-continued



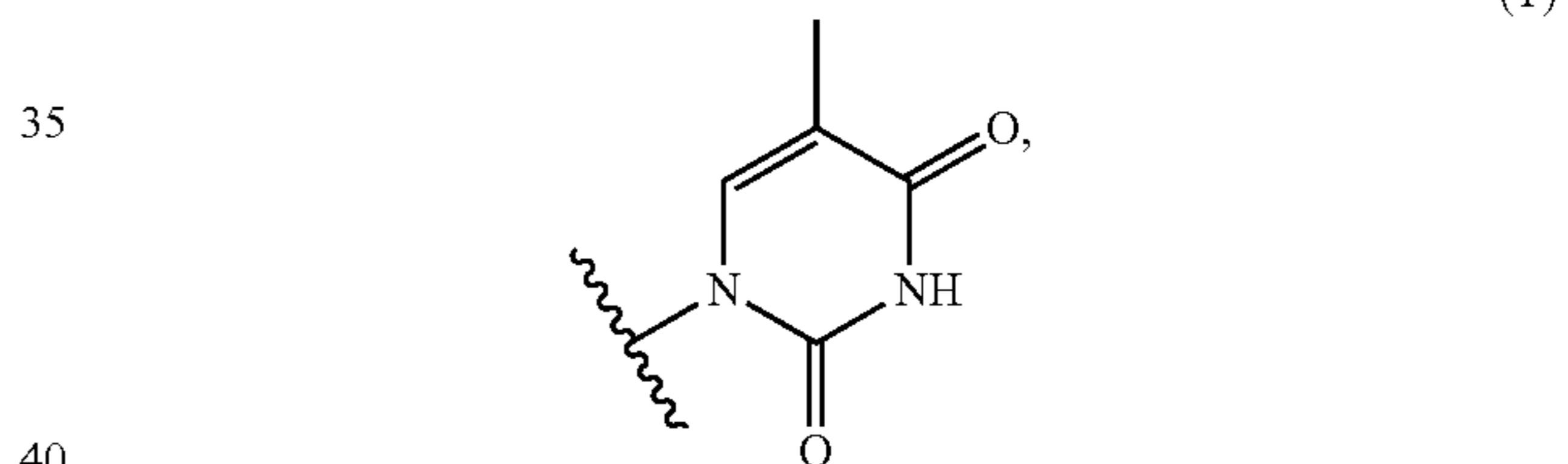
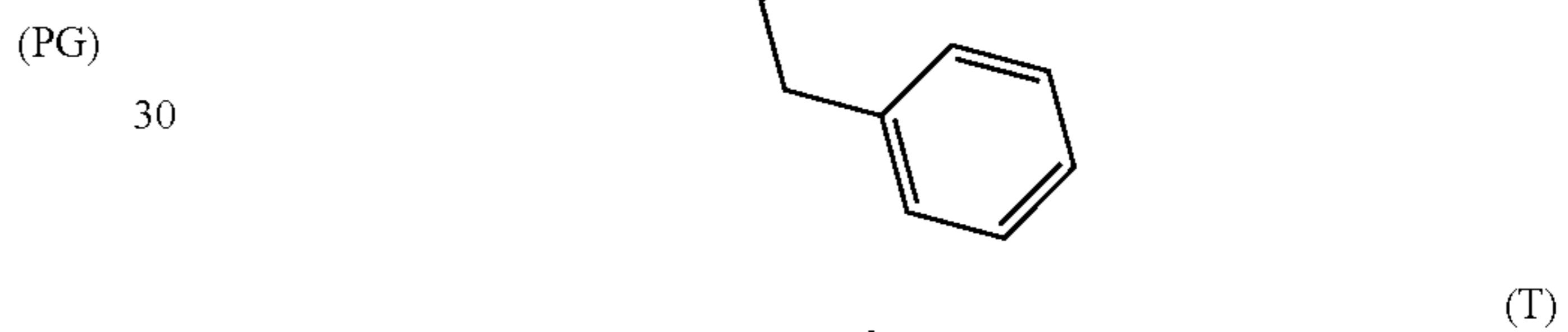
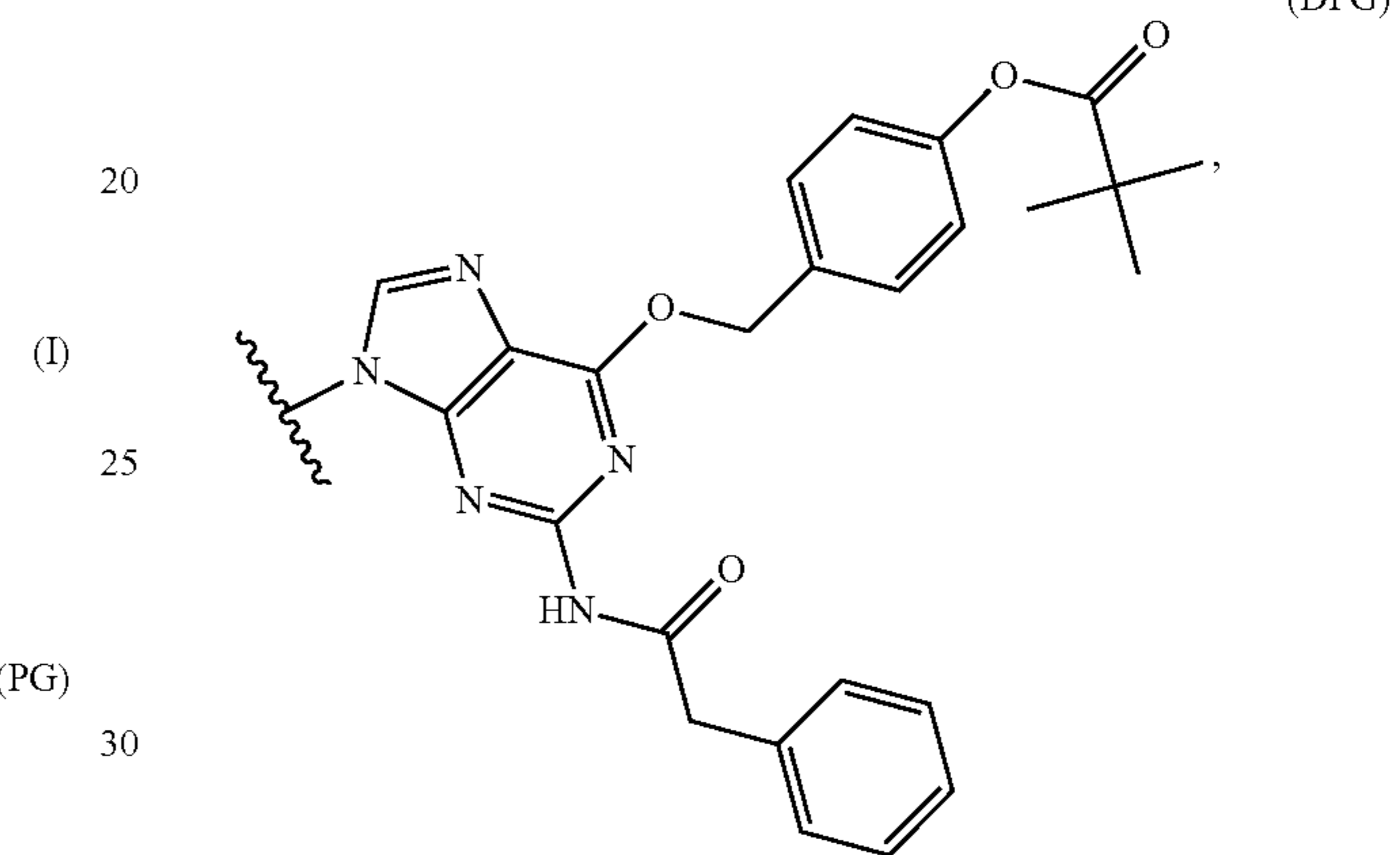
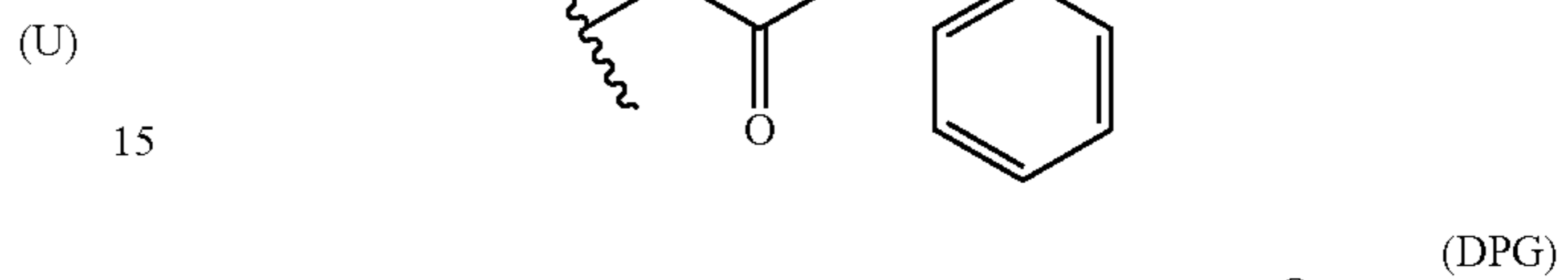
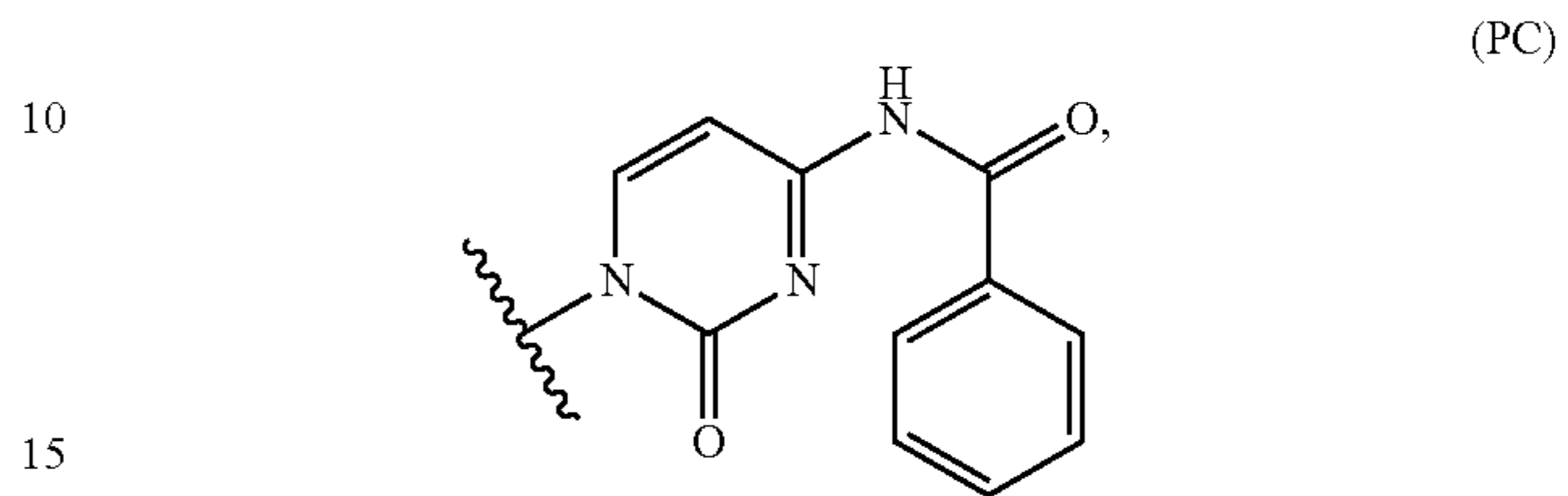
and



104

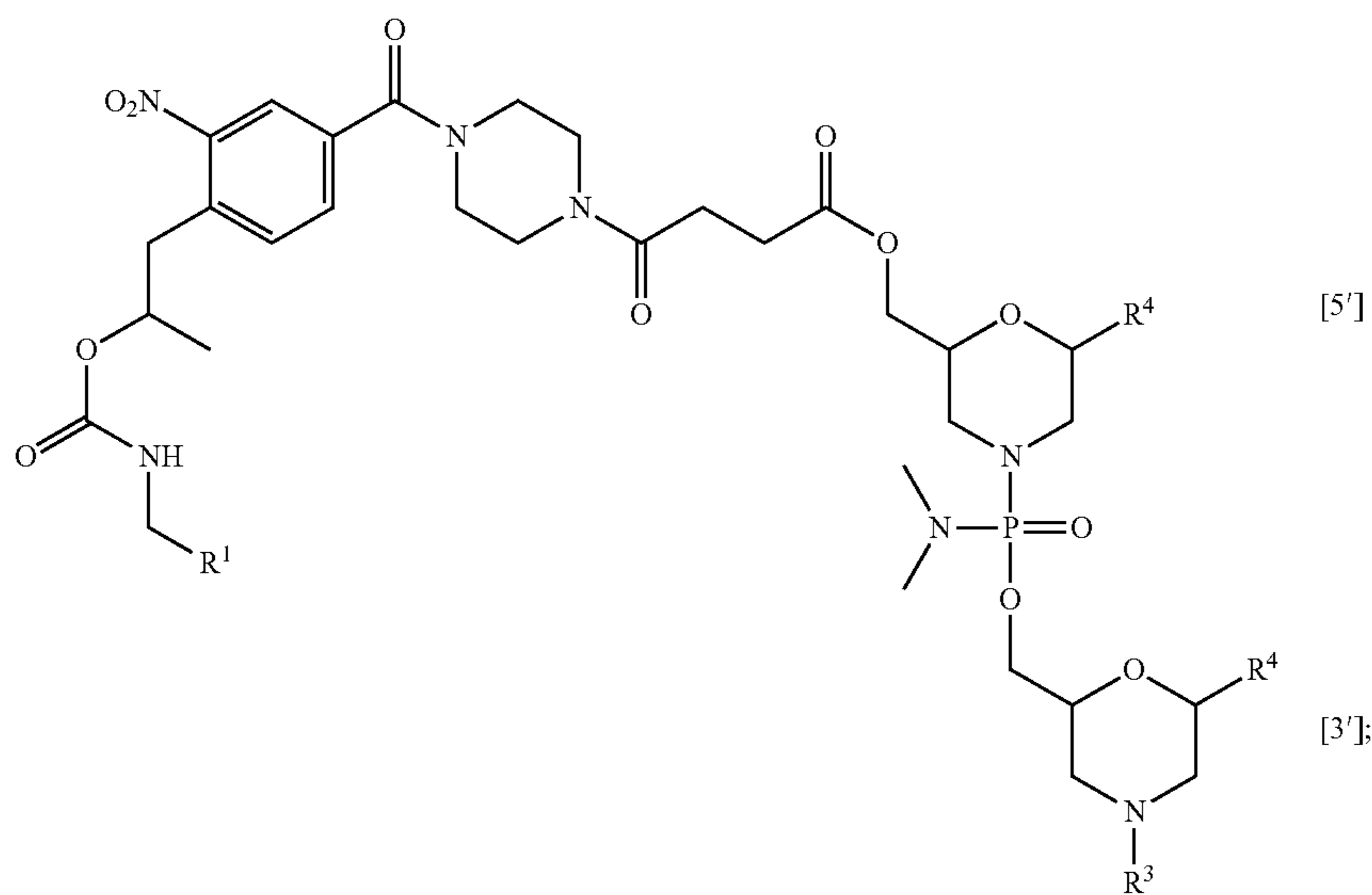
wherein R¹ is a support-medium, R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

R⁴, independently at each occurrence, is selected from the group consisting of:



to form a compound of Formula (S5);

(S5)

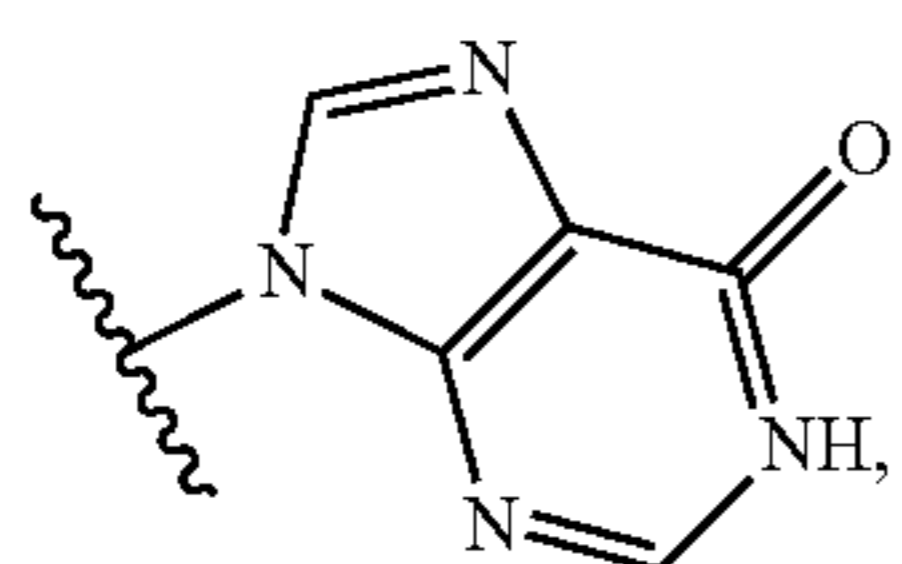
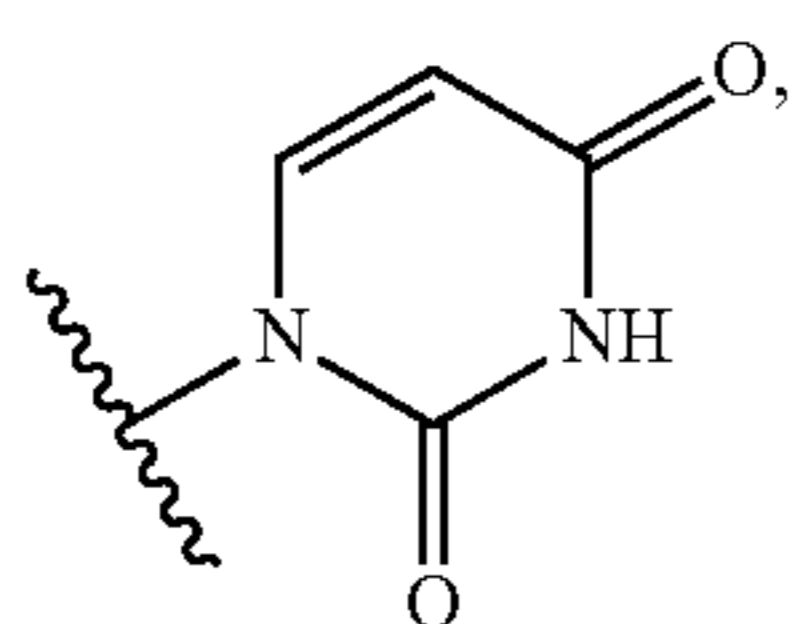
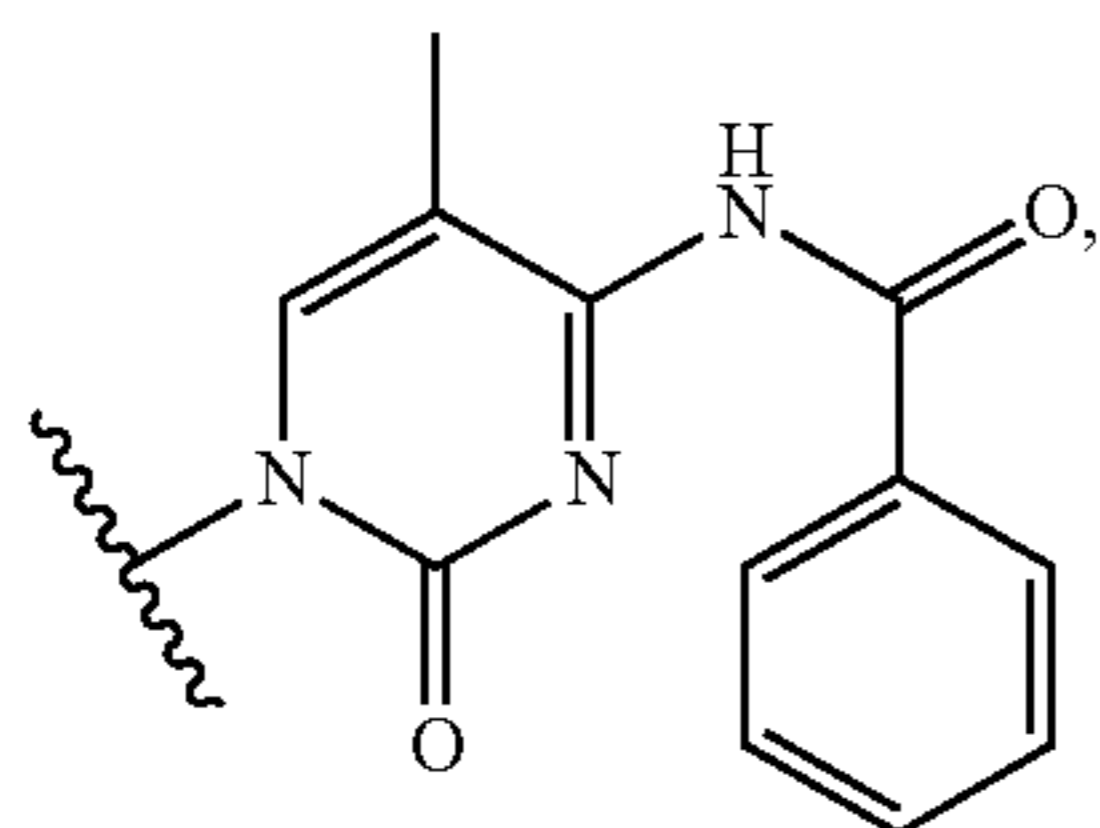
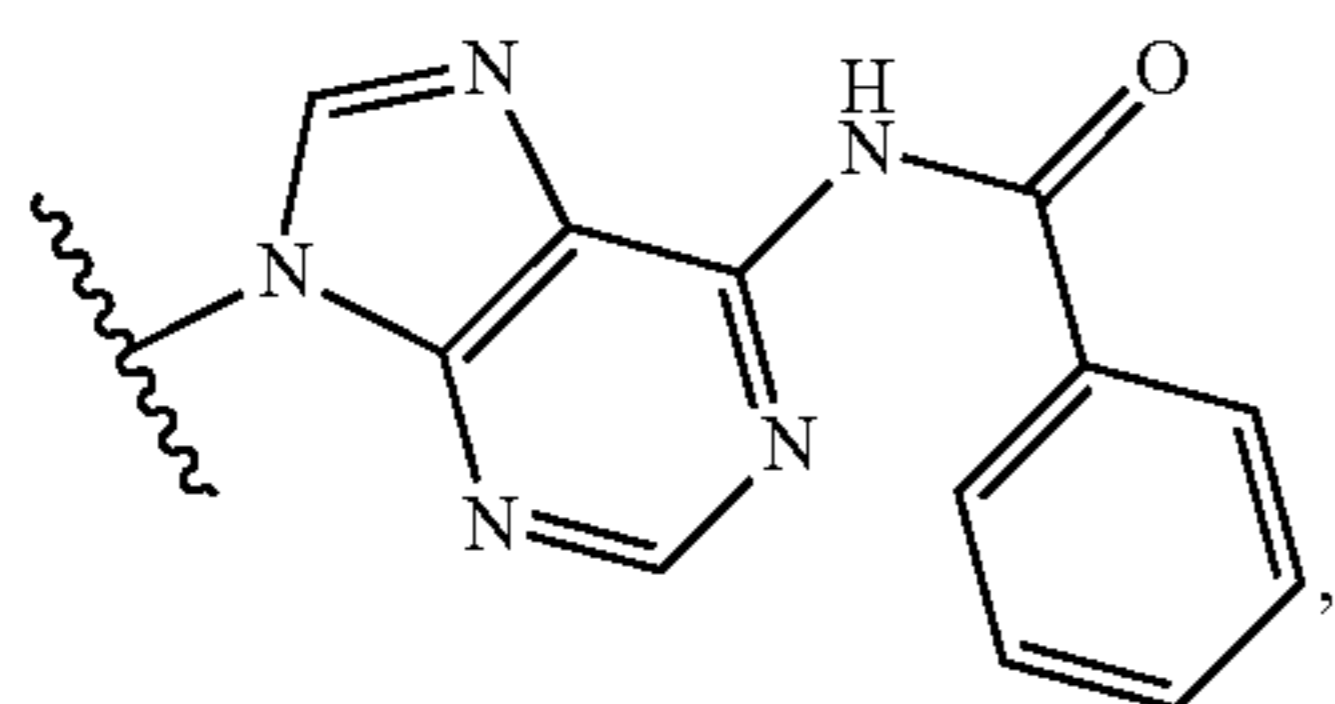


[5']

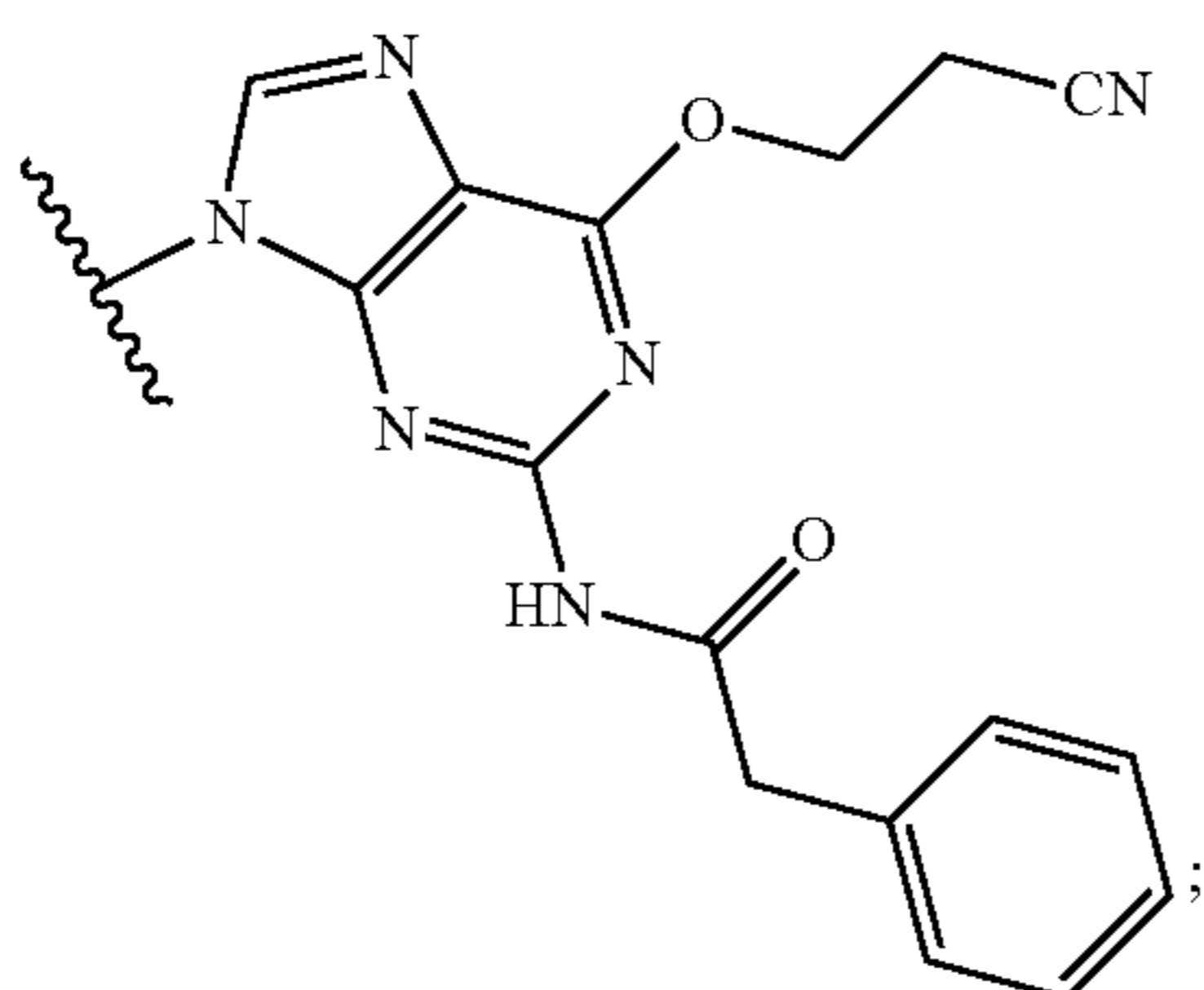
[3']

105

-continued



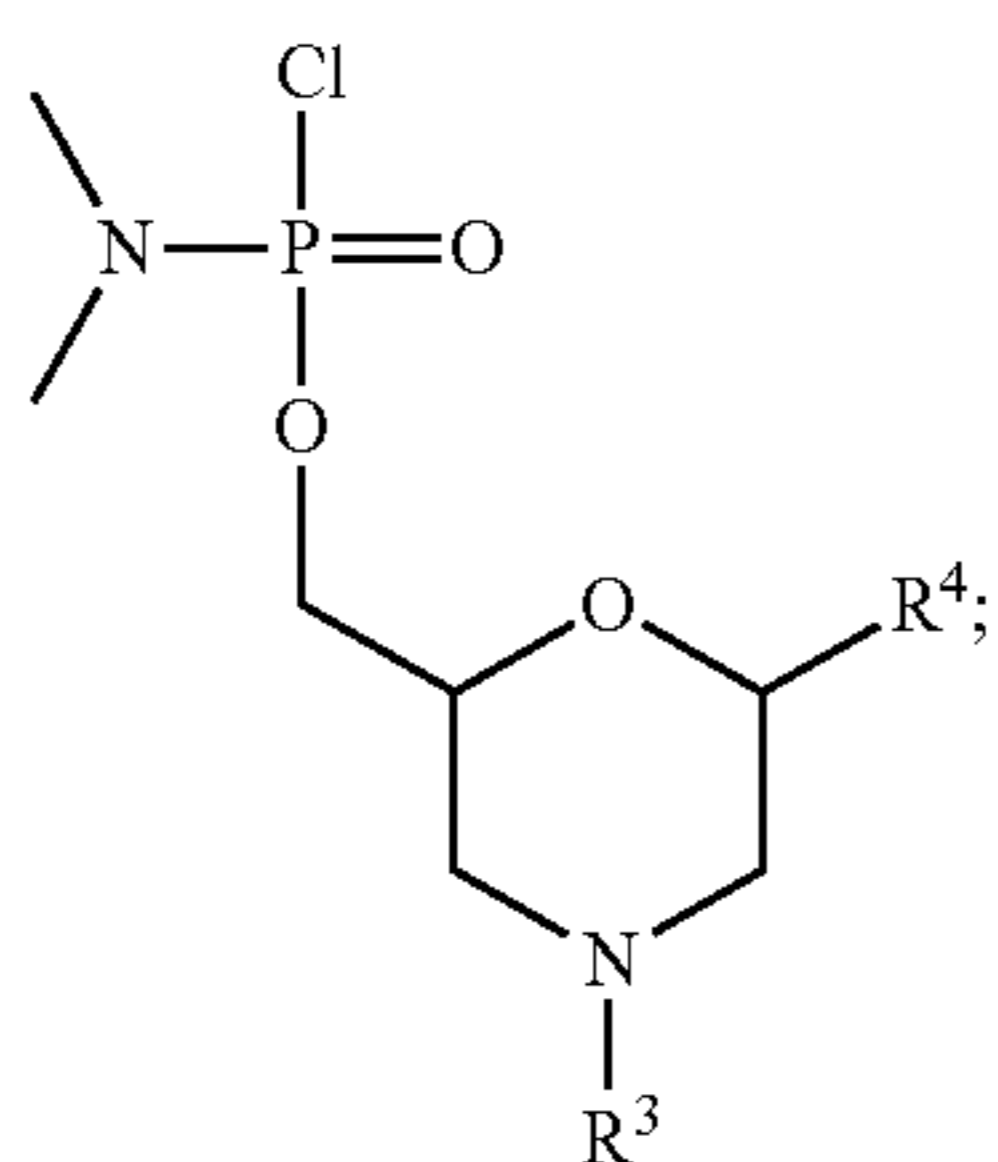
and



(d) performing n-2 iterations of the sequential steps of:

(d1) contacting the product formed by the immediately prior step with a deblocking agent; and

(d2) contacting the compound formed by the immediately prior step with a compound of Formula (A8):

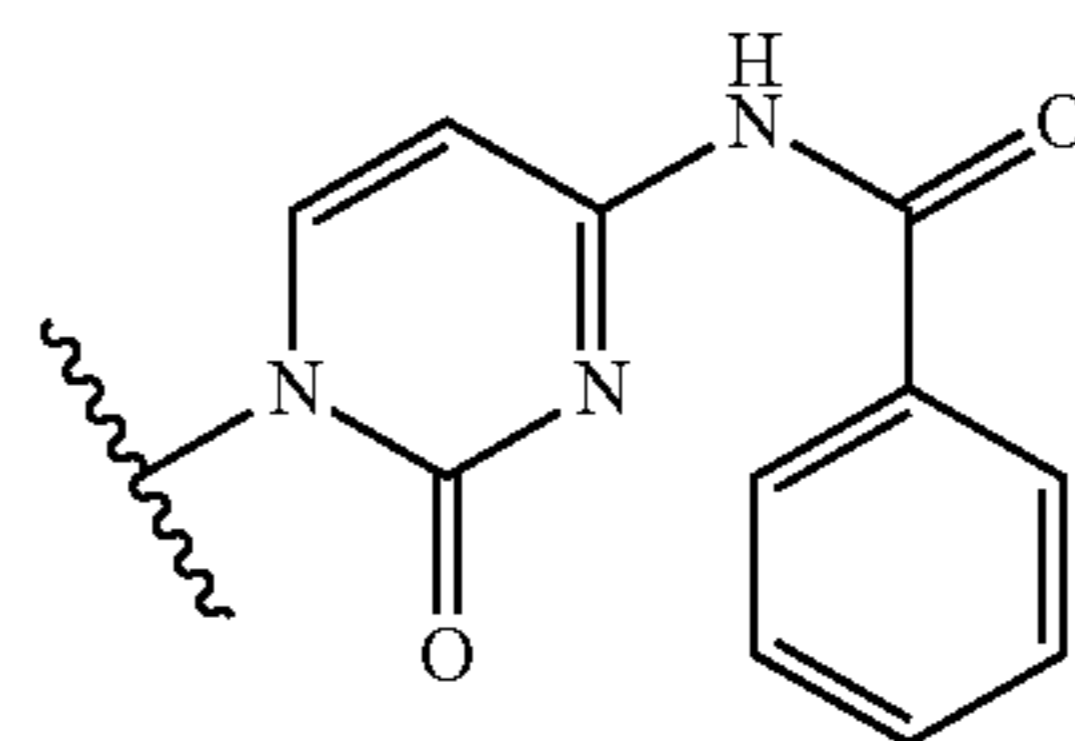


wherein R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each compound of Formula (A8), selected from the group consisting of:

106

(PA)

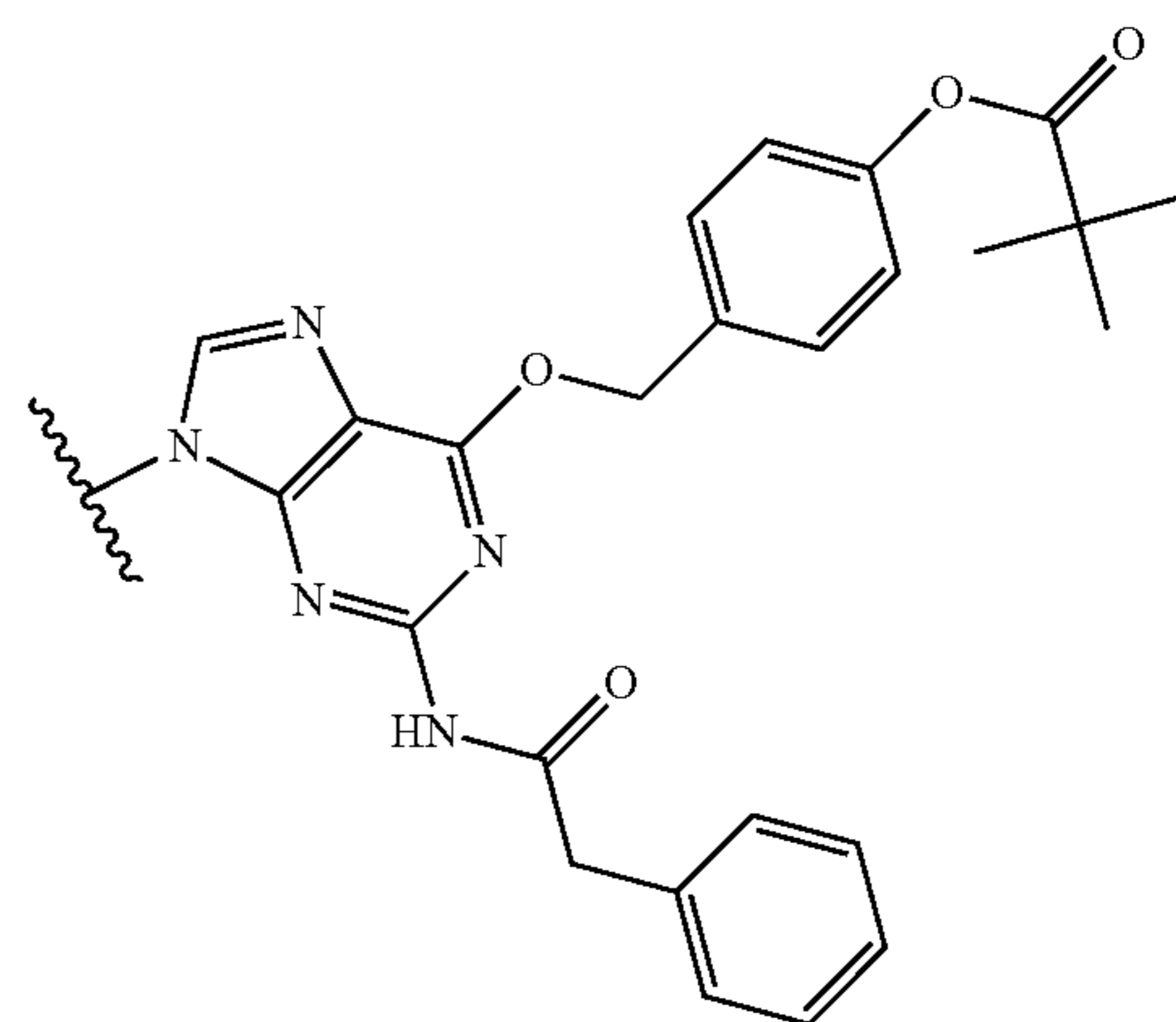
5



(PC)

(P5mC)

10



(DPG)

(U)

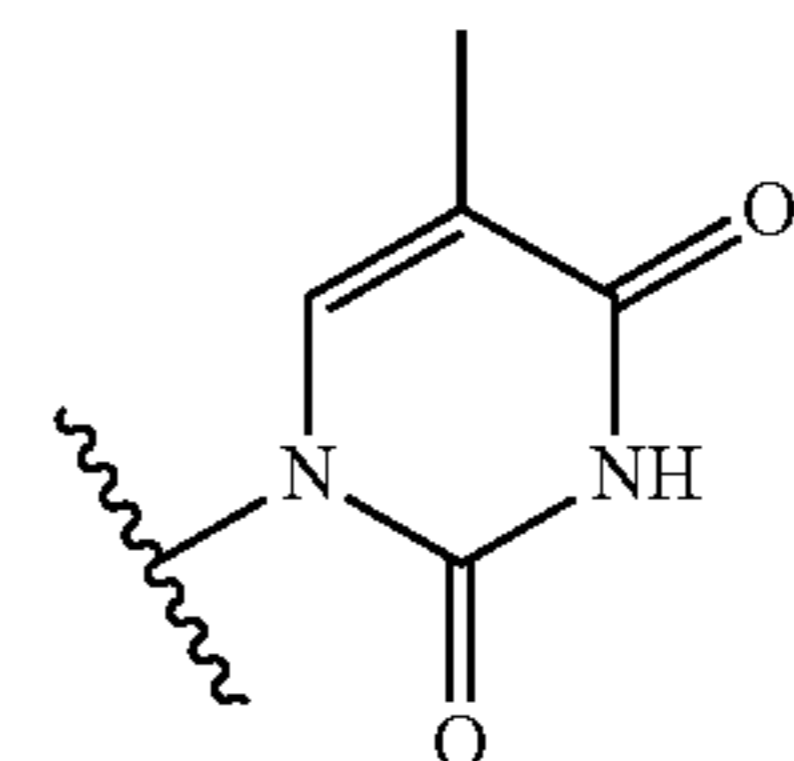
20

(I)

25

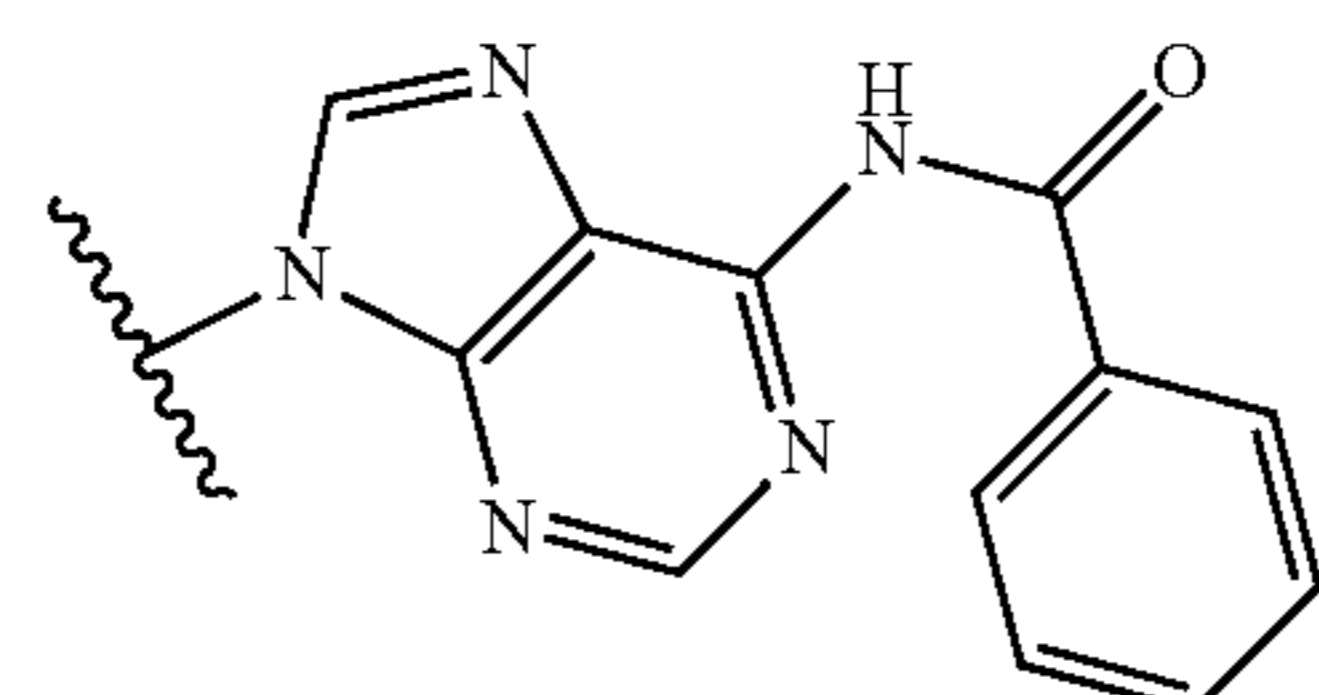
(PG)

30



(T)

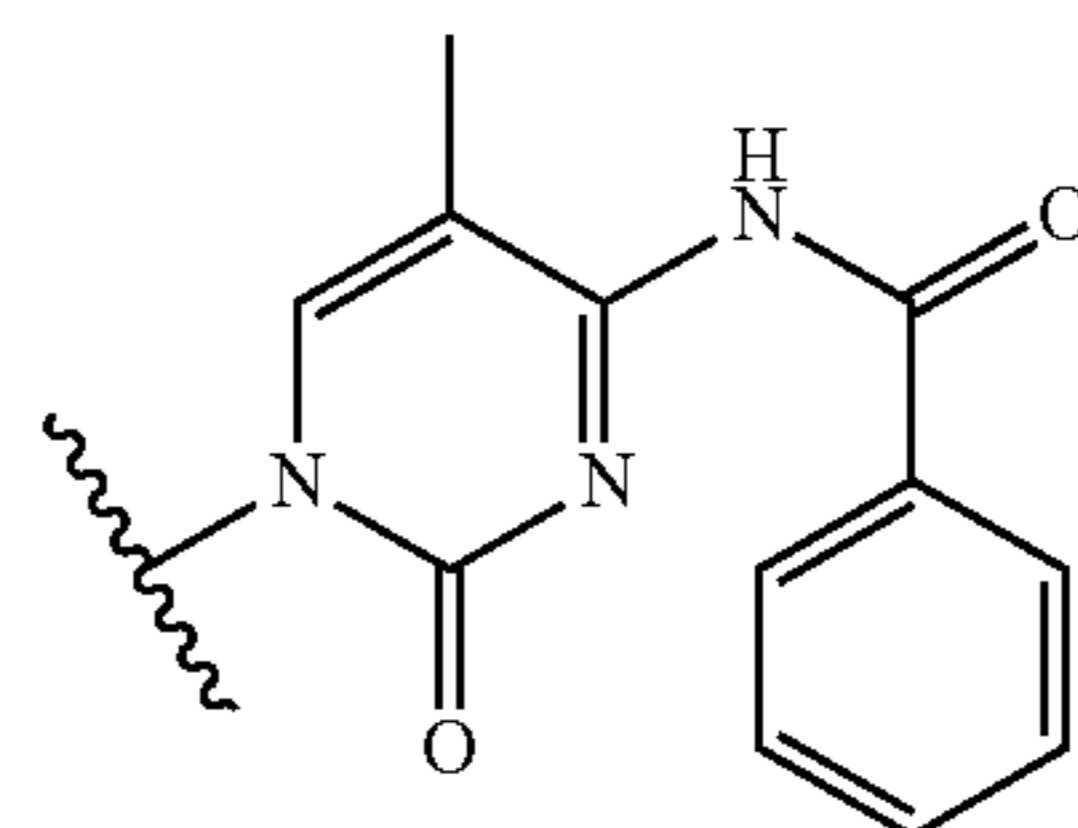
35



(PA)

40

(P5mC)

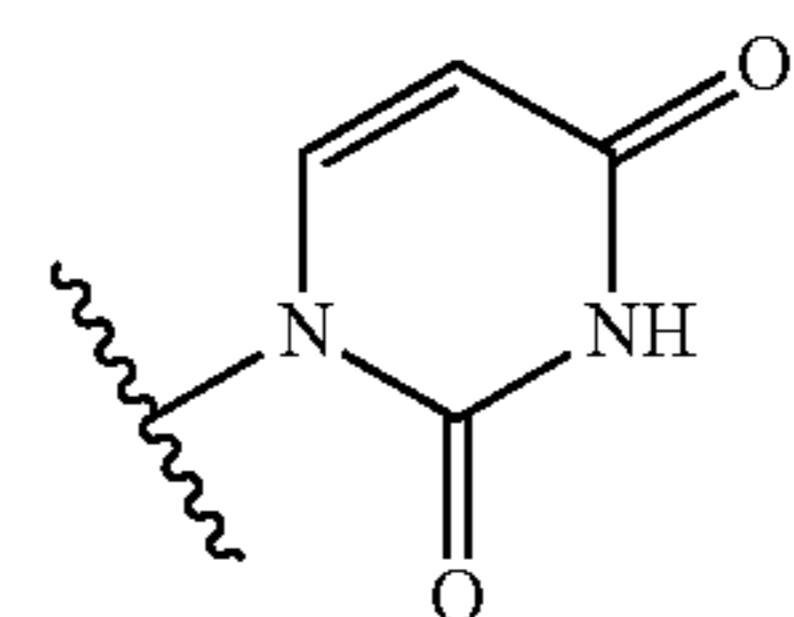


50

(A8)

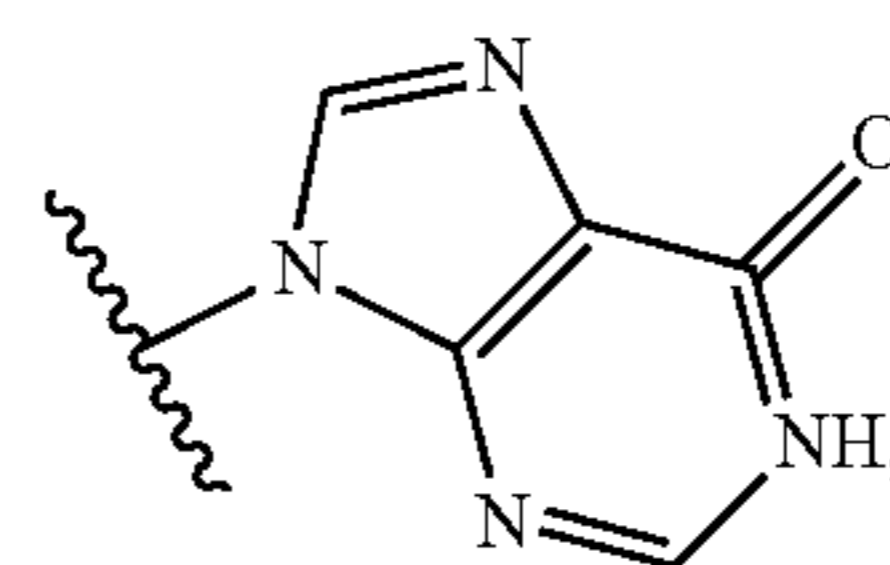
(U)

55



60

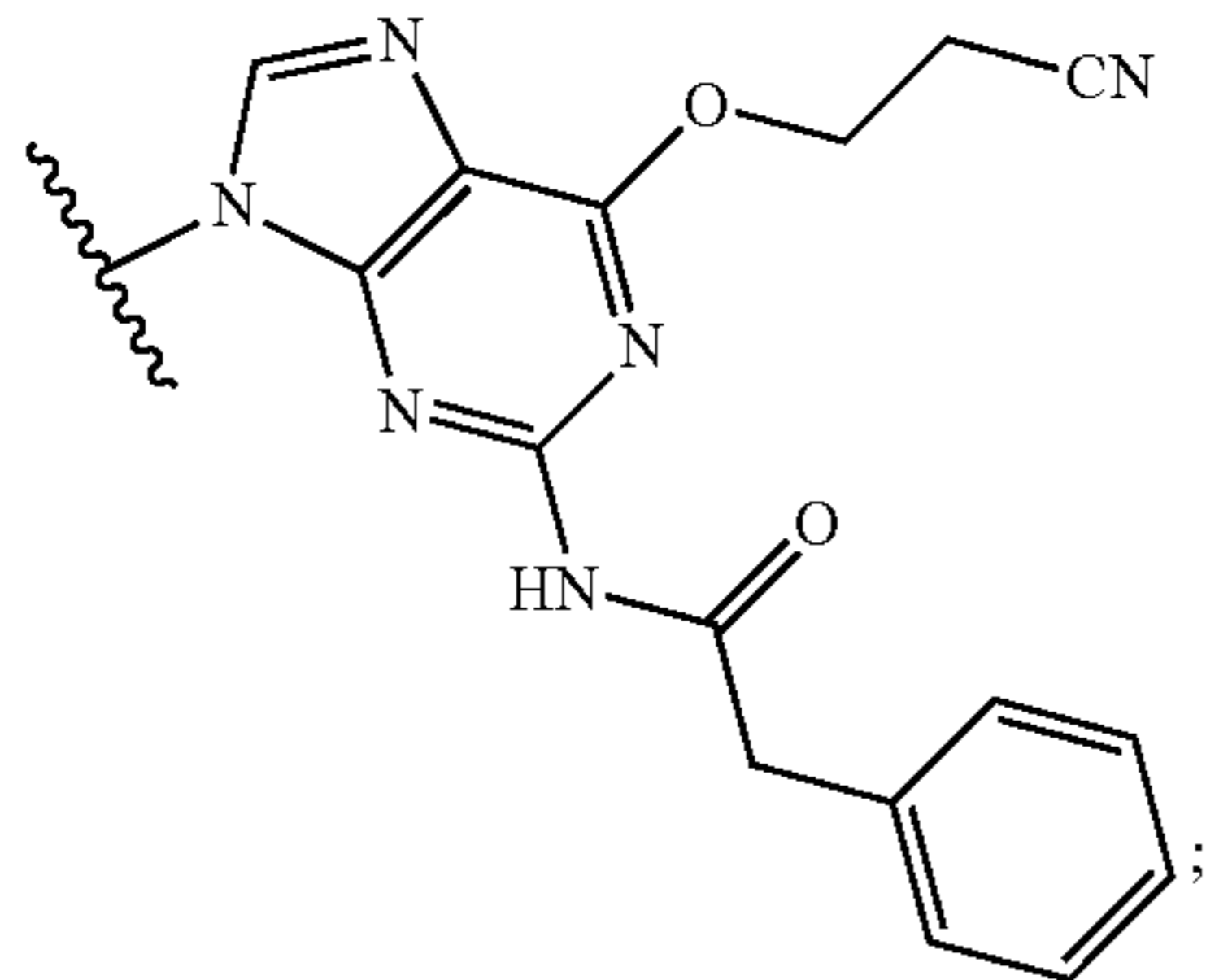
(I)



and

107

-continued



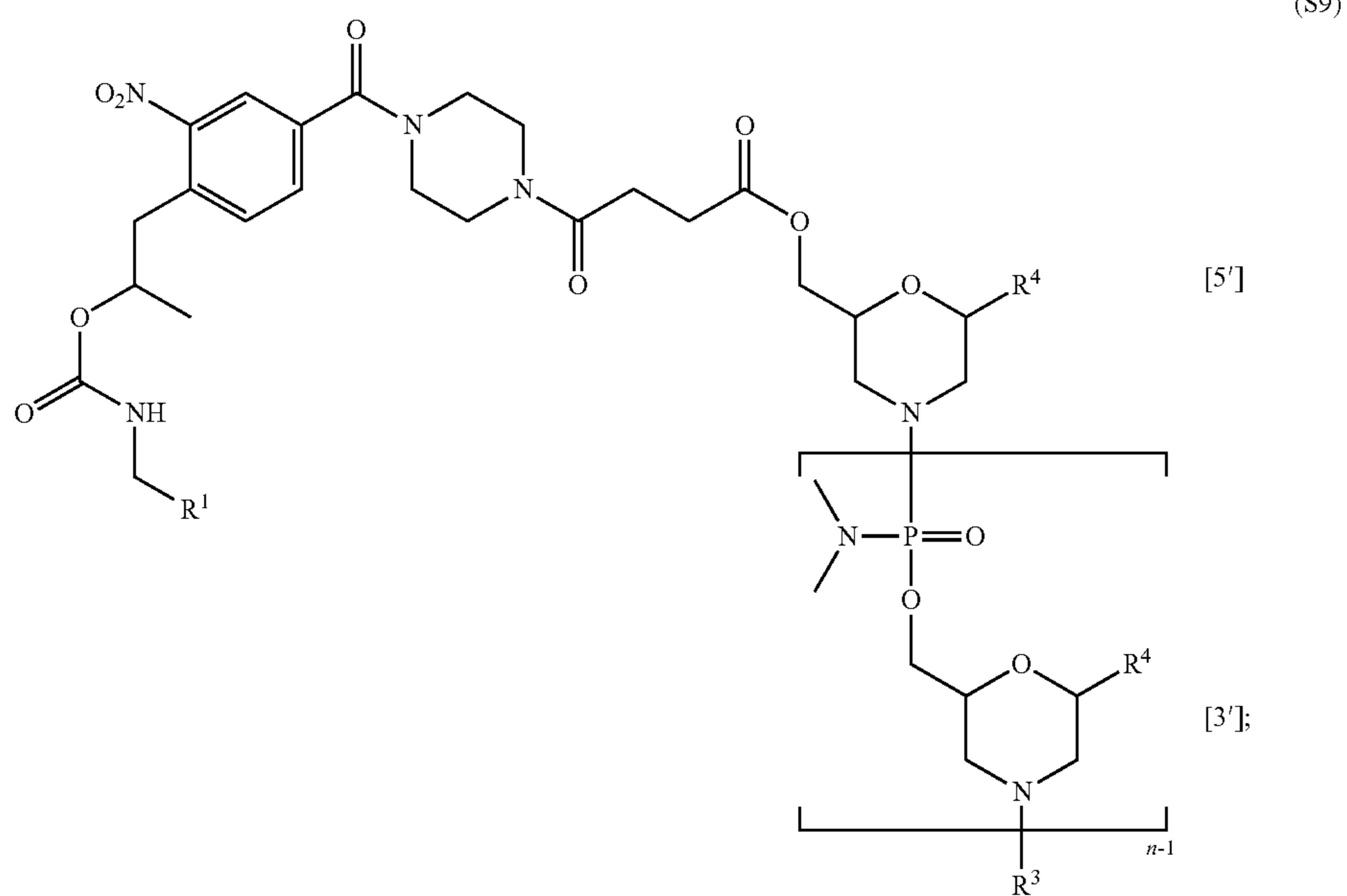
(PG)

5

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108

to form a compound of Formula (S9):

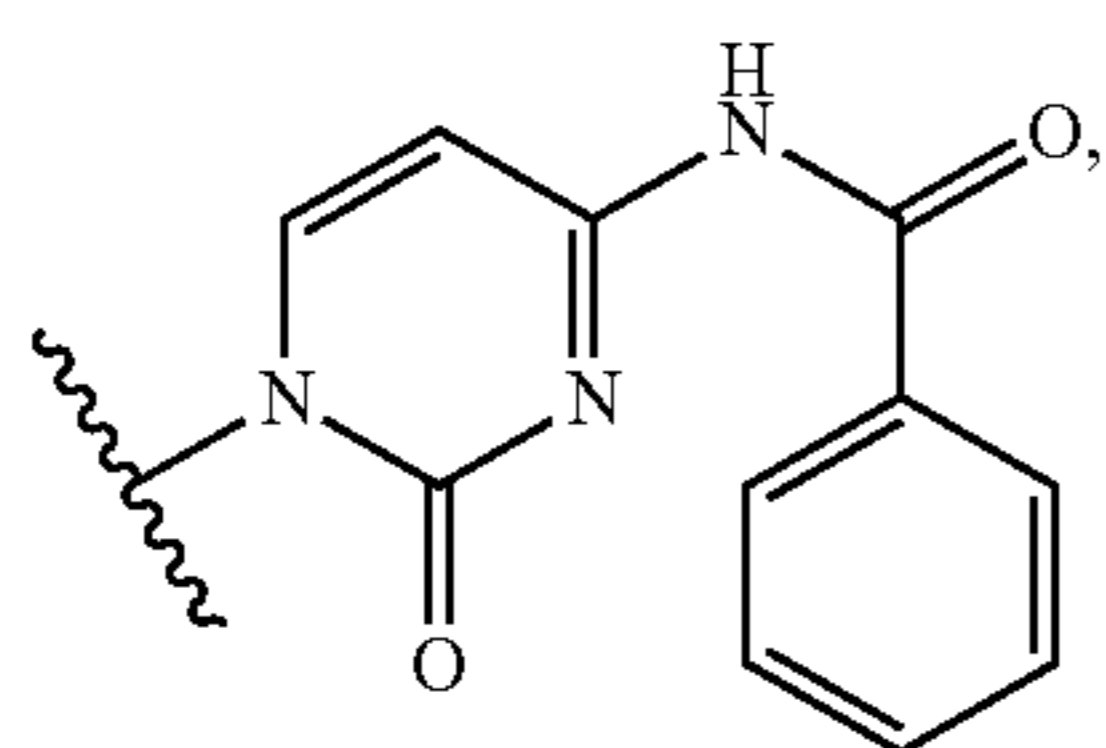


(S9)

[5']

[3']

wherein n is an integer from 10 to 40, R^1 is a support-medium, R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R^4 is, independently for each occurrence, selected from the group consisting of:



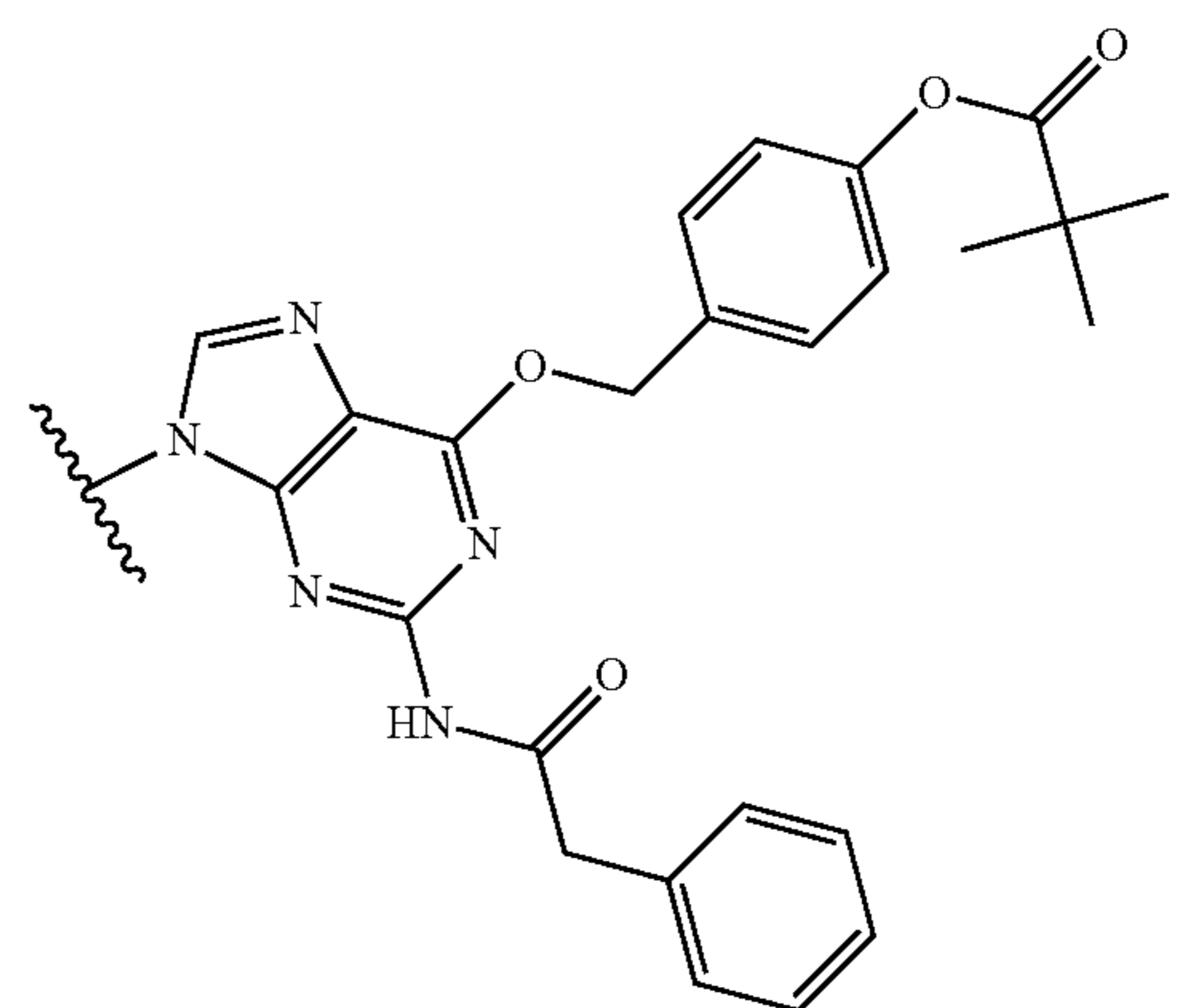
-continued

(PC)

55

60

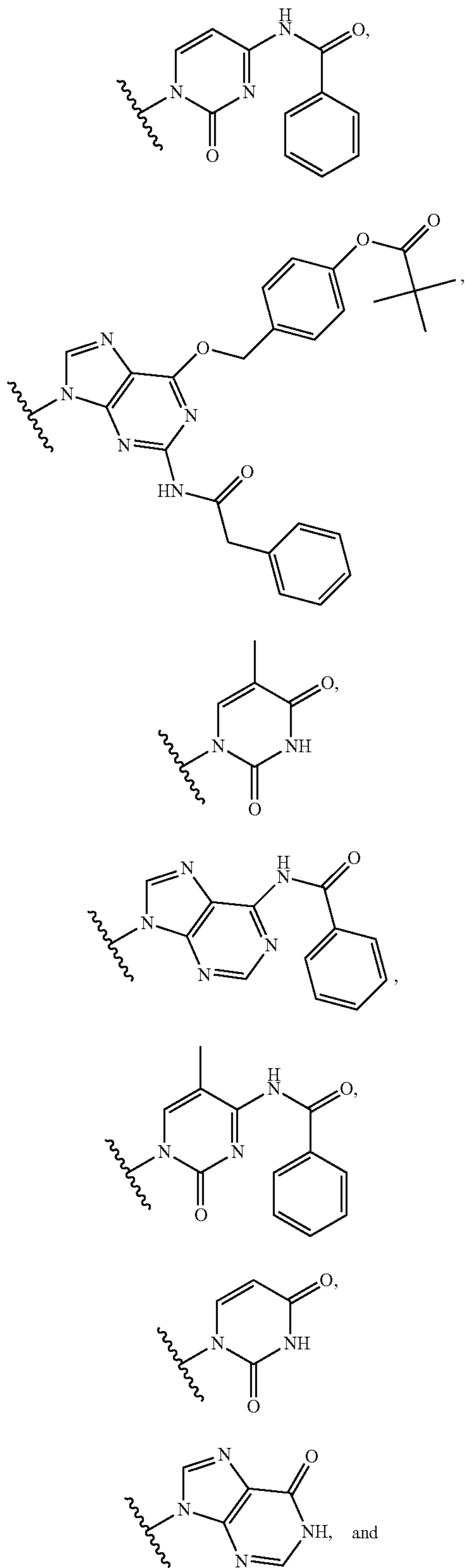
65



(DPG)

111

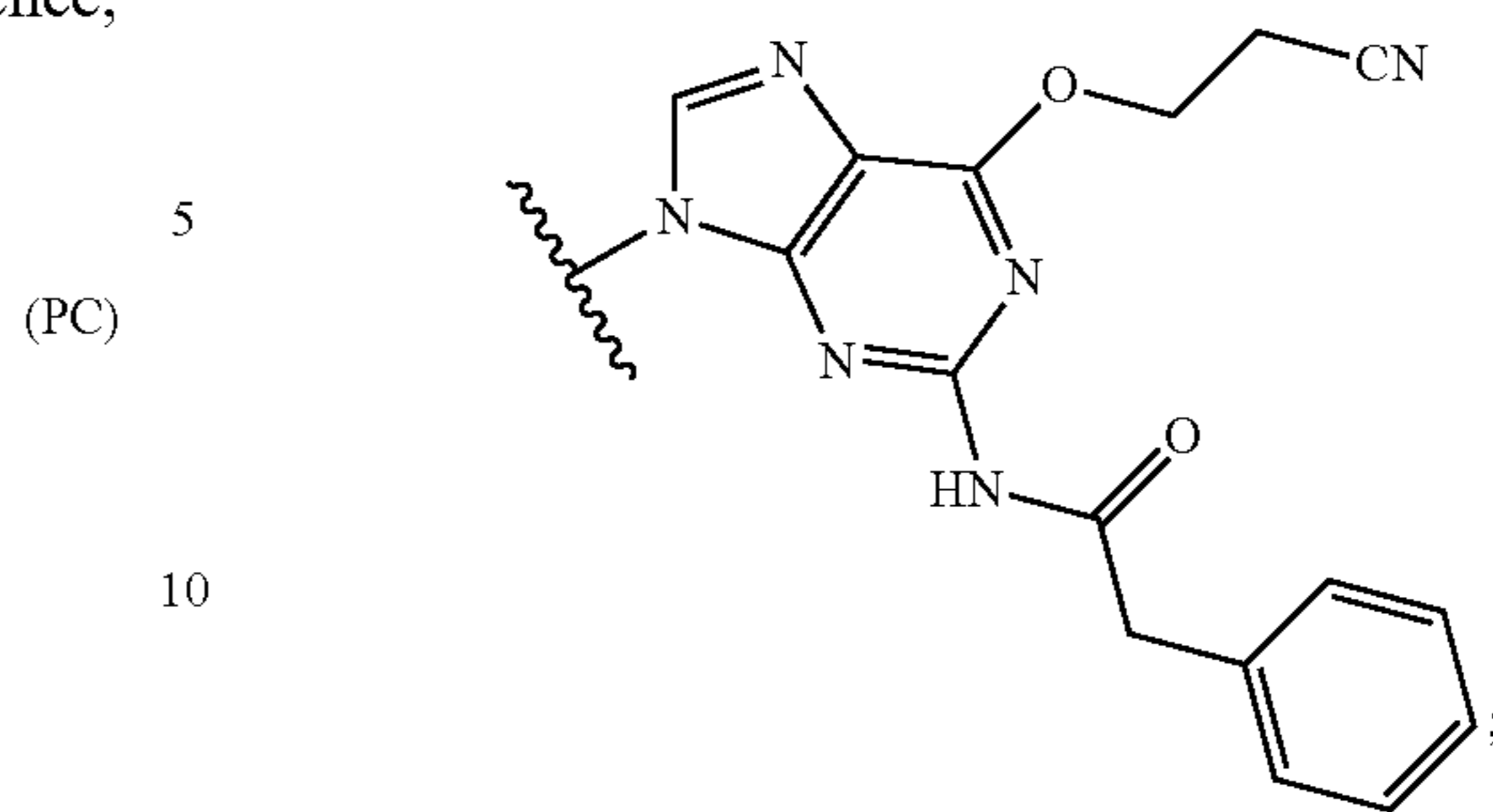
wherein n is an integer from 10 to 40, R¹ is a support-medium, and R⁴ is, independently for each occurrence, selected from the group consisting of:



112

-continued

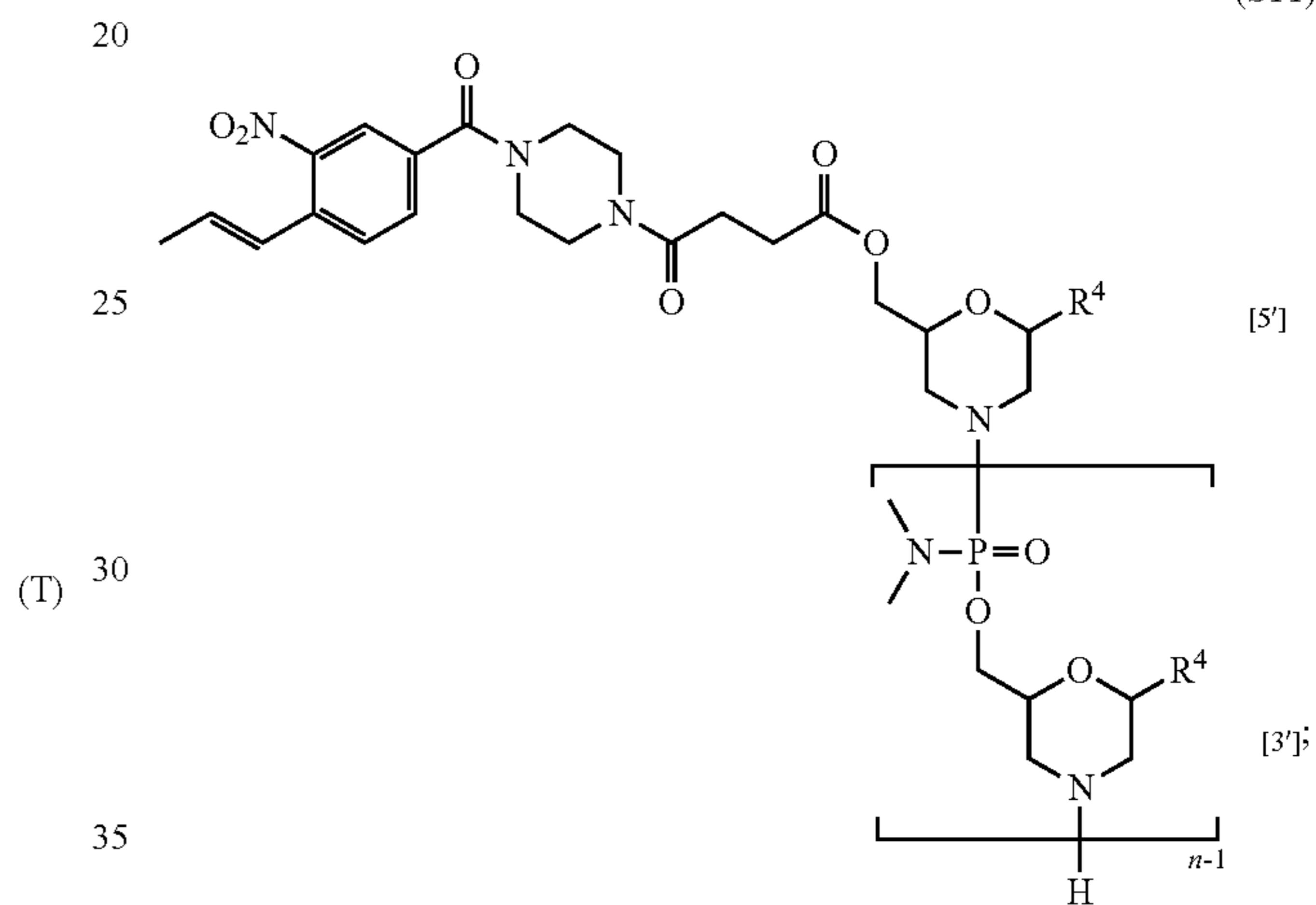
(PG)



(DPG)

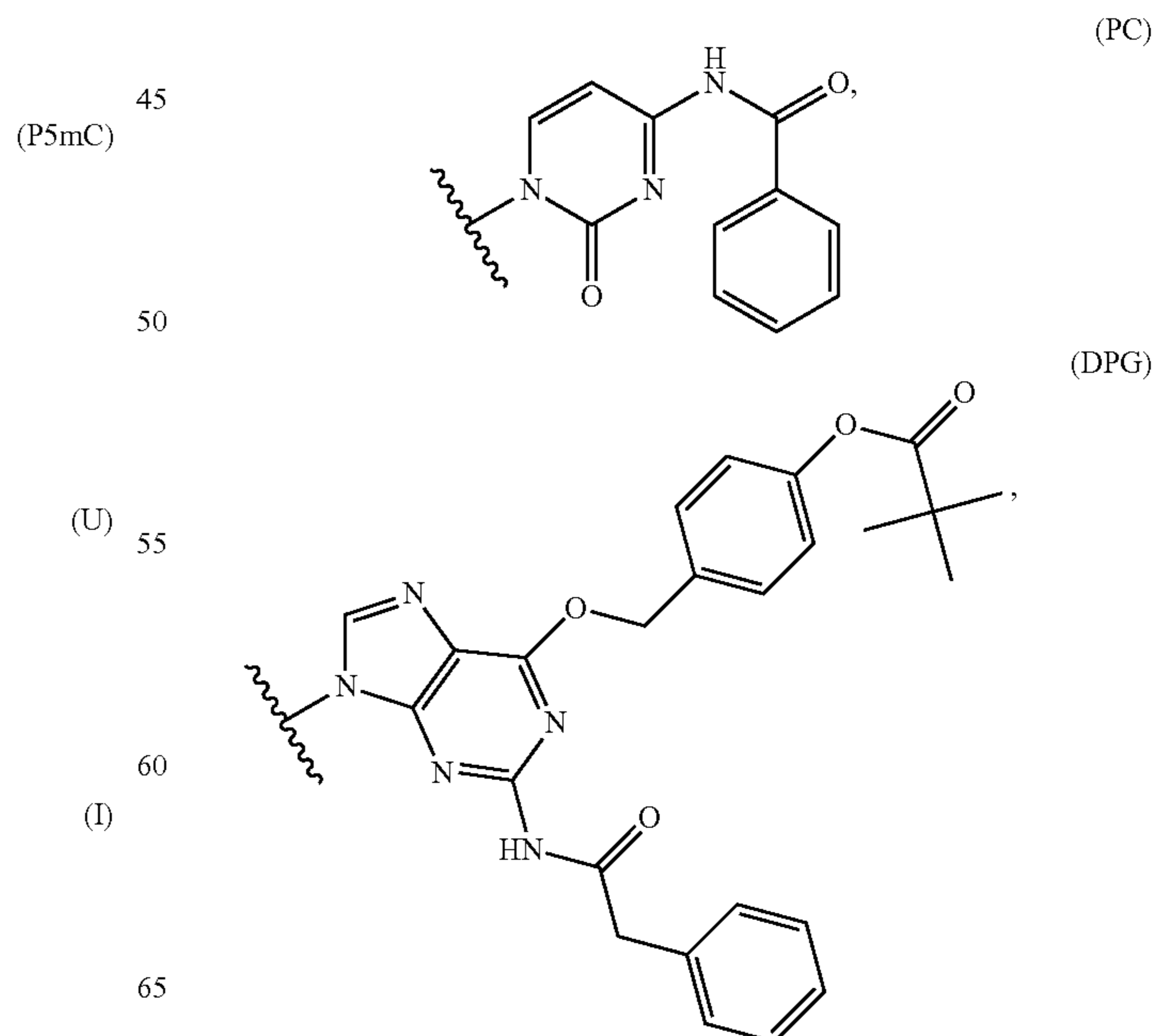
15 (f) contacting the compound of Formula (S10) with a cleaving agent to form a compound of Formula (S11):

(S11)



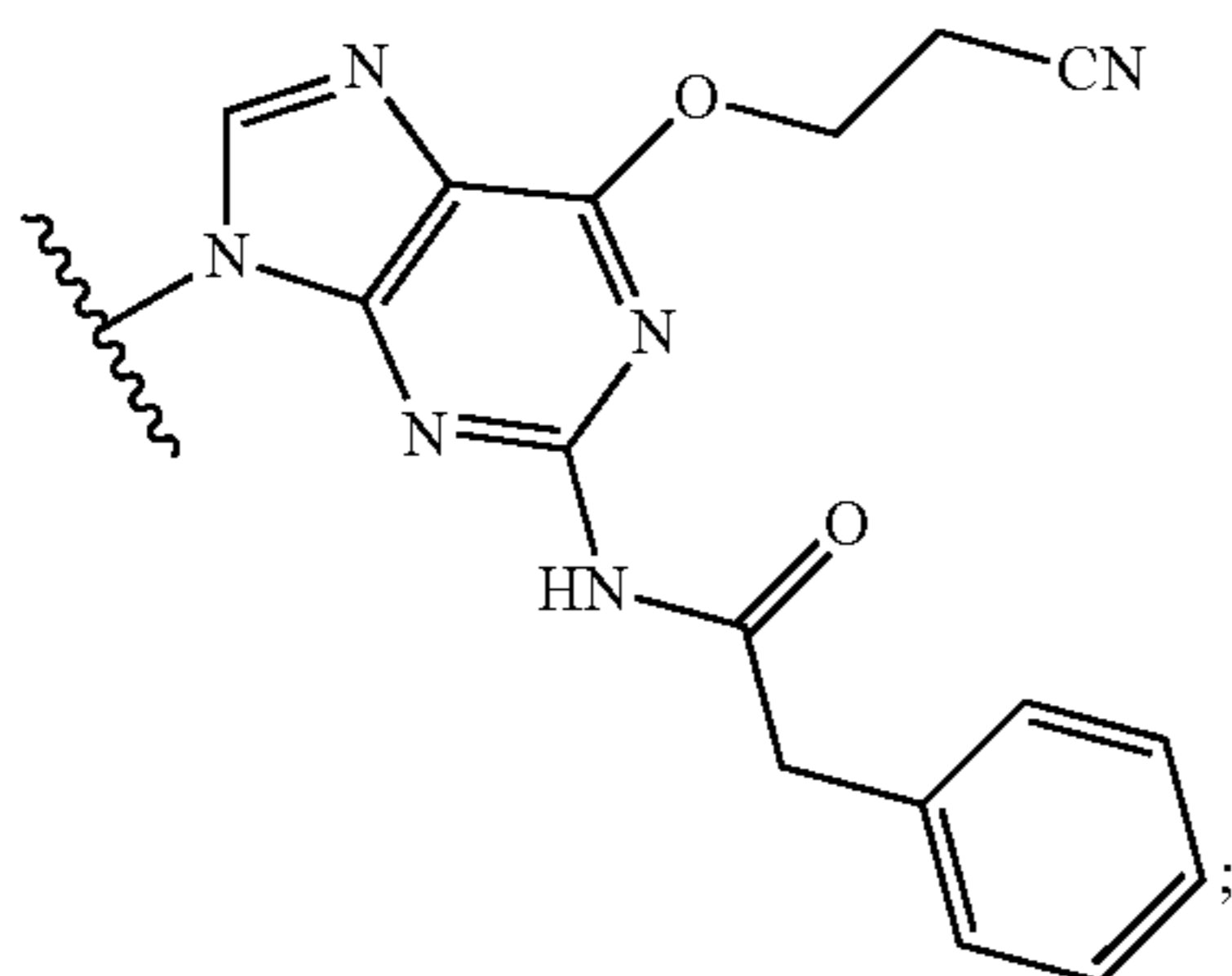
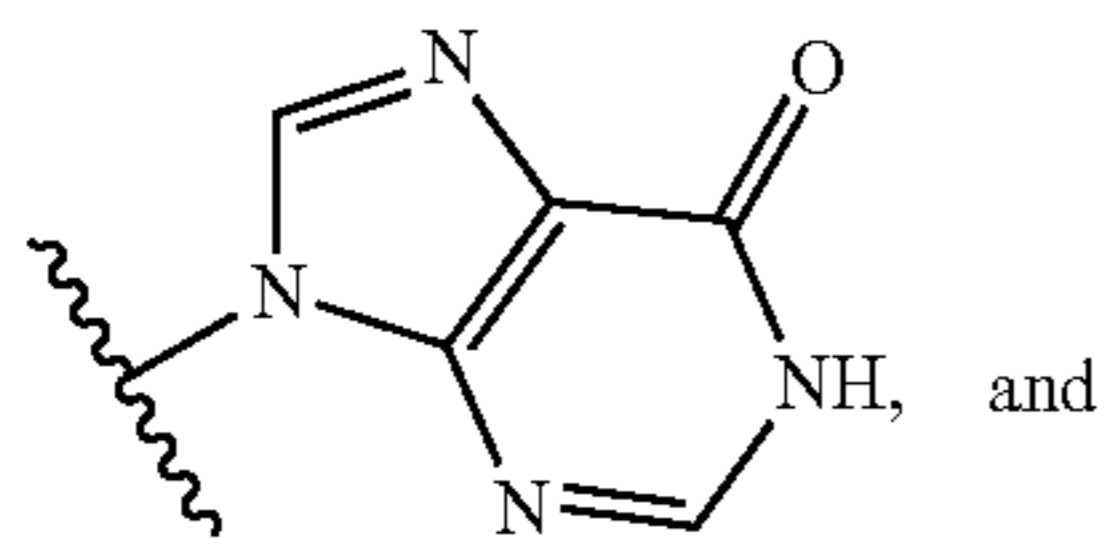
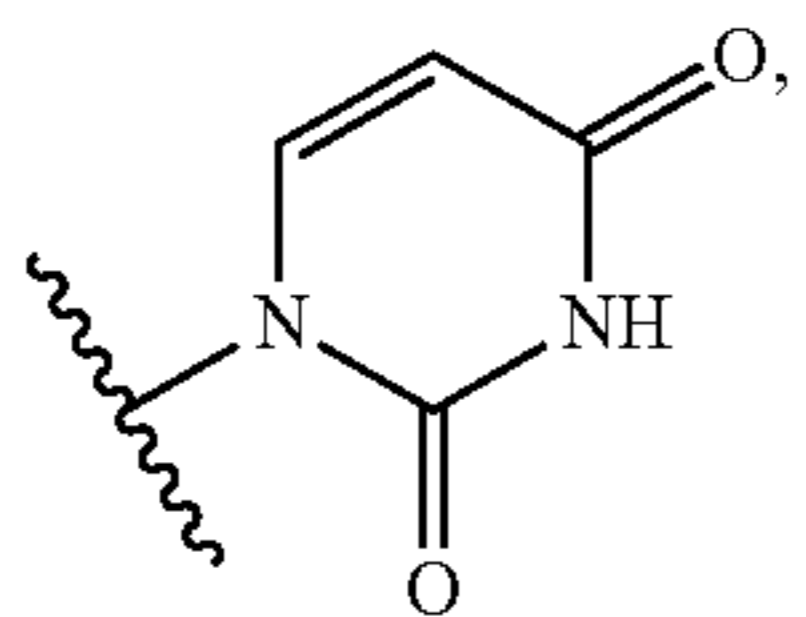
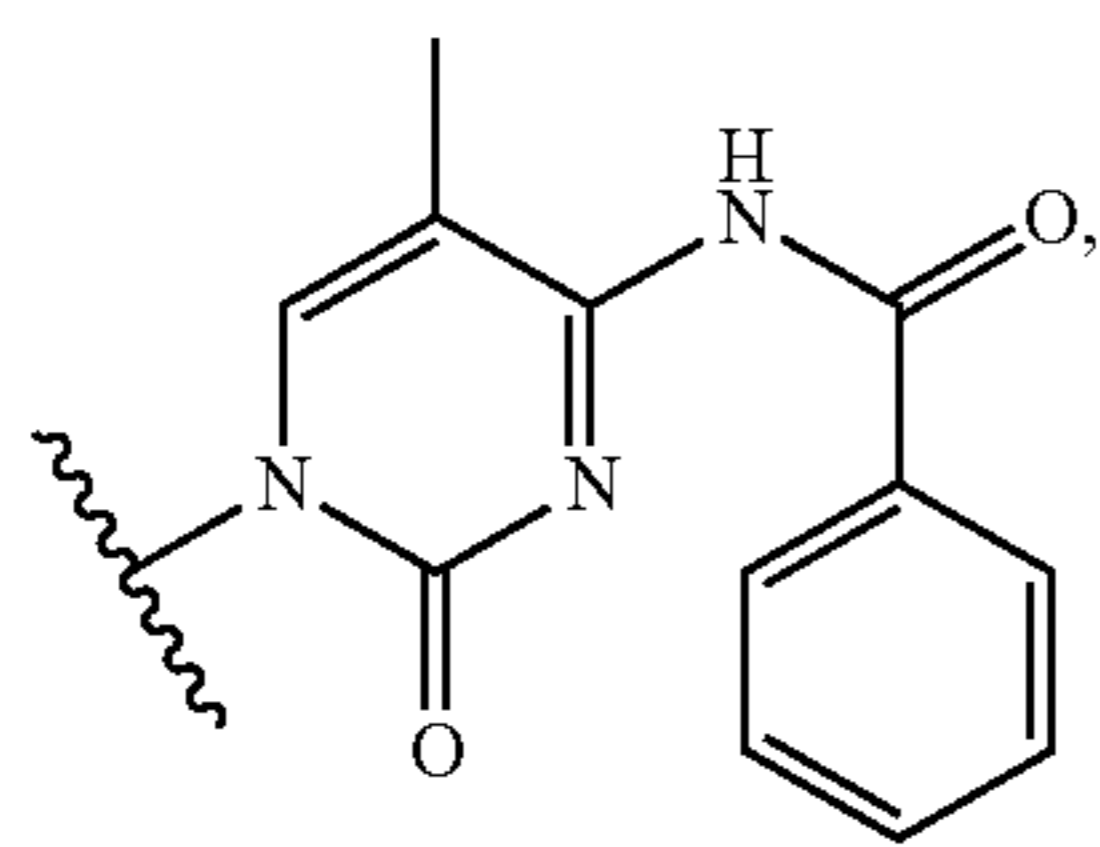
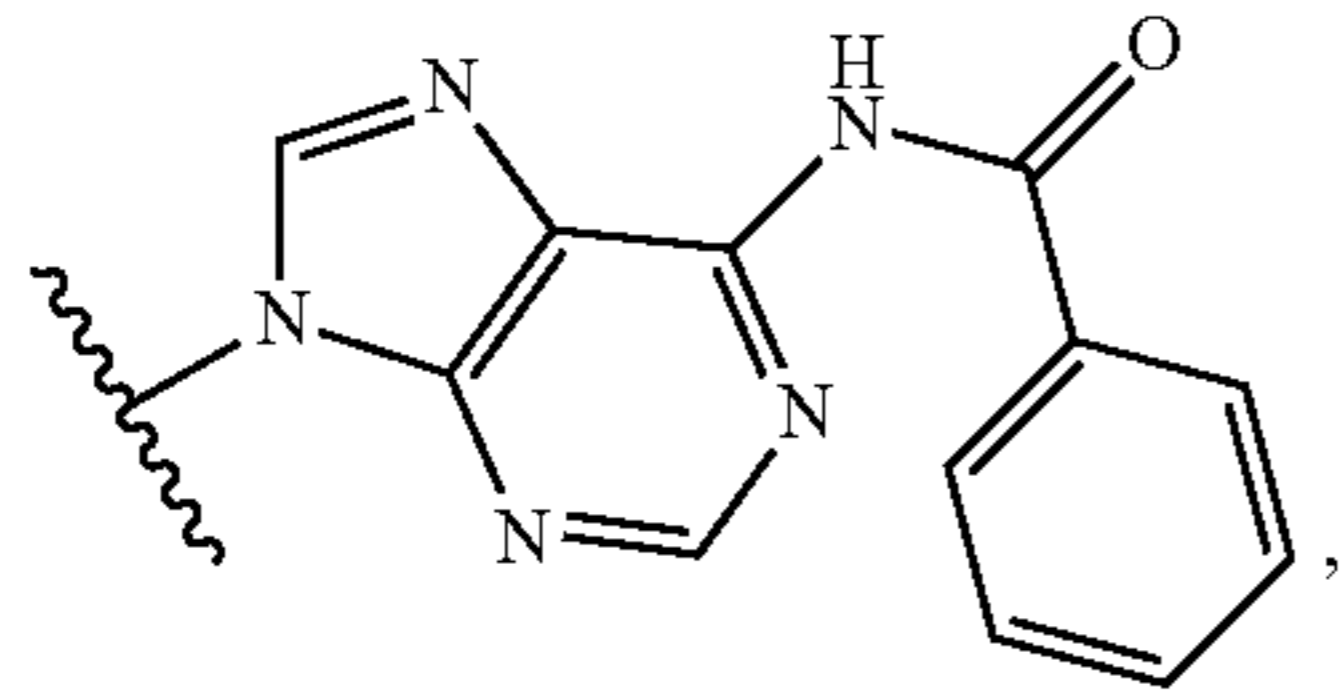
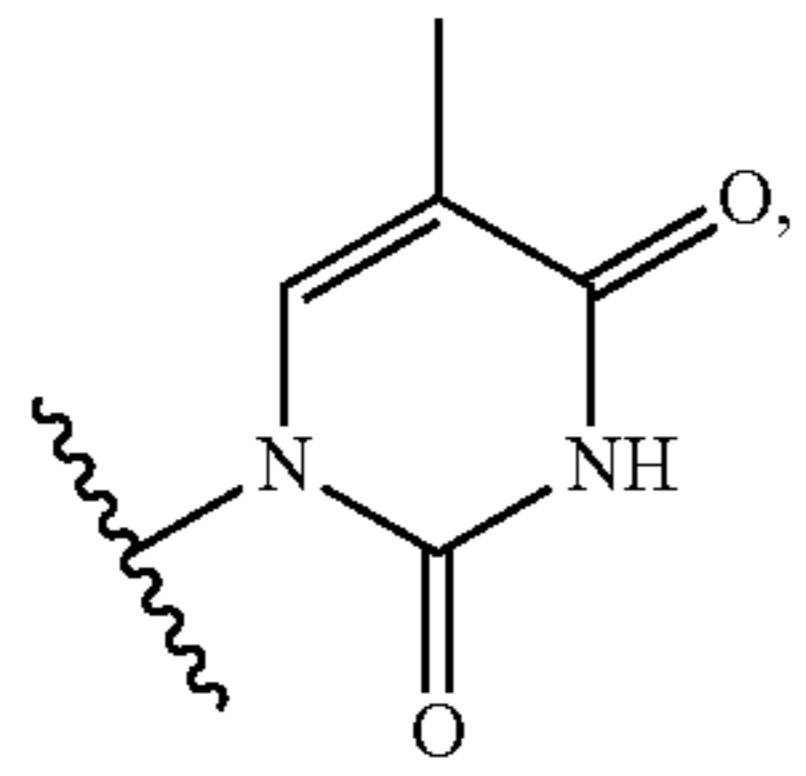
(PA)

40 wherein n is an integer from 10 to 40, and R⁴ is, independently for each occurrence, selected from the group consisting of:



113

-continued



114

and

(T)

(g) contacting the compound of Formula (S11) with a deprotecting agent to form the oligomeric compound of Formula (S).

Processes for Preparing Eteplirsen

(PA)

Provided herein are processes for preparing Eteplirsen.

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In another aspect, provided herein is a process for preparing an oligomeric compound of Formula (E):

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(P5mC)

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(U)

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(I)

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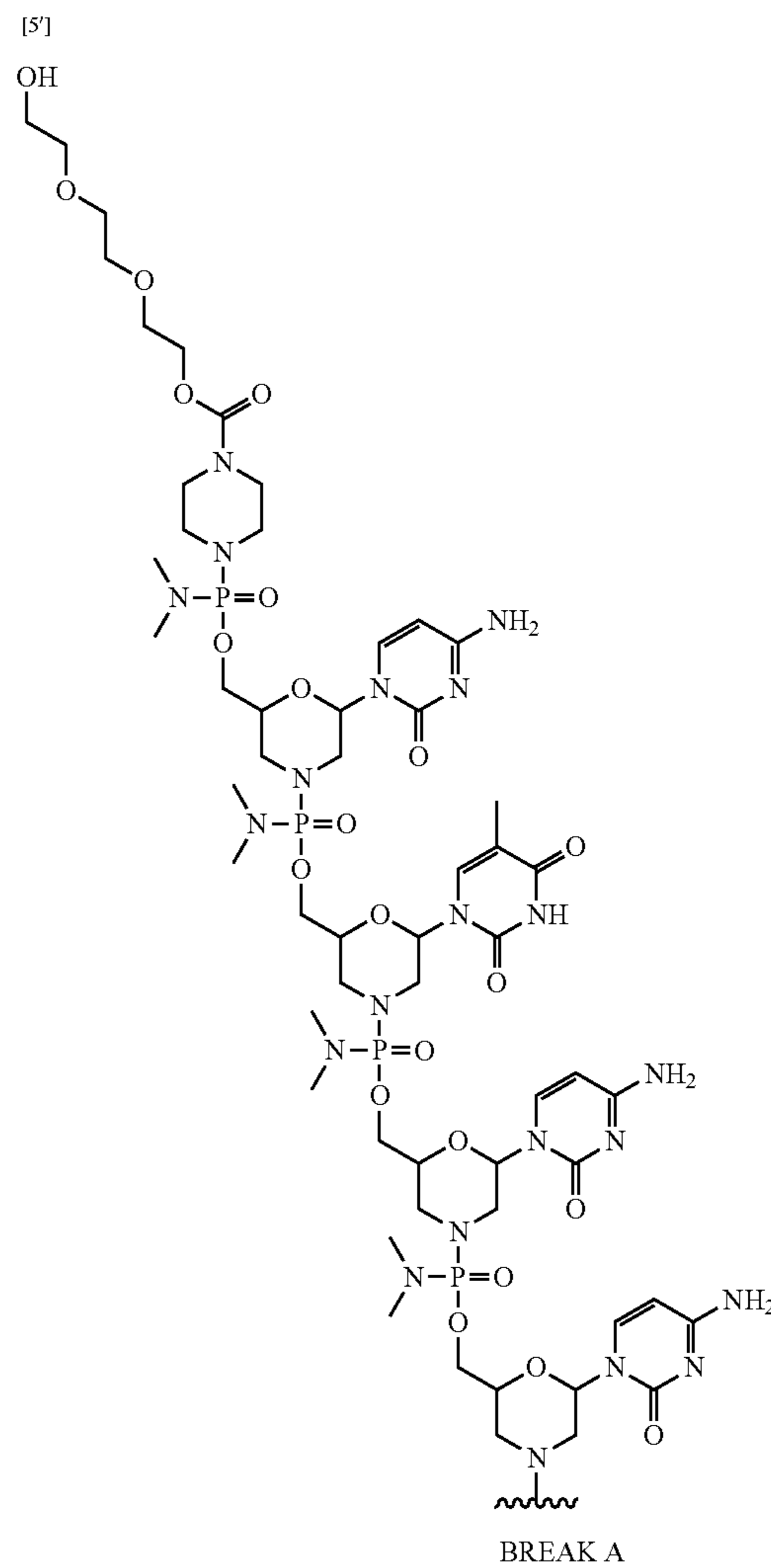
(PG)

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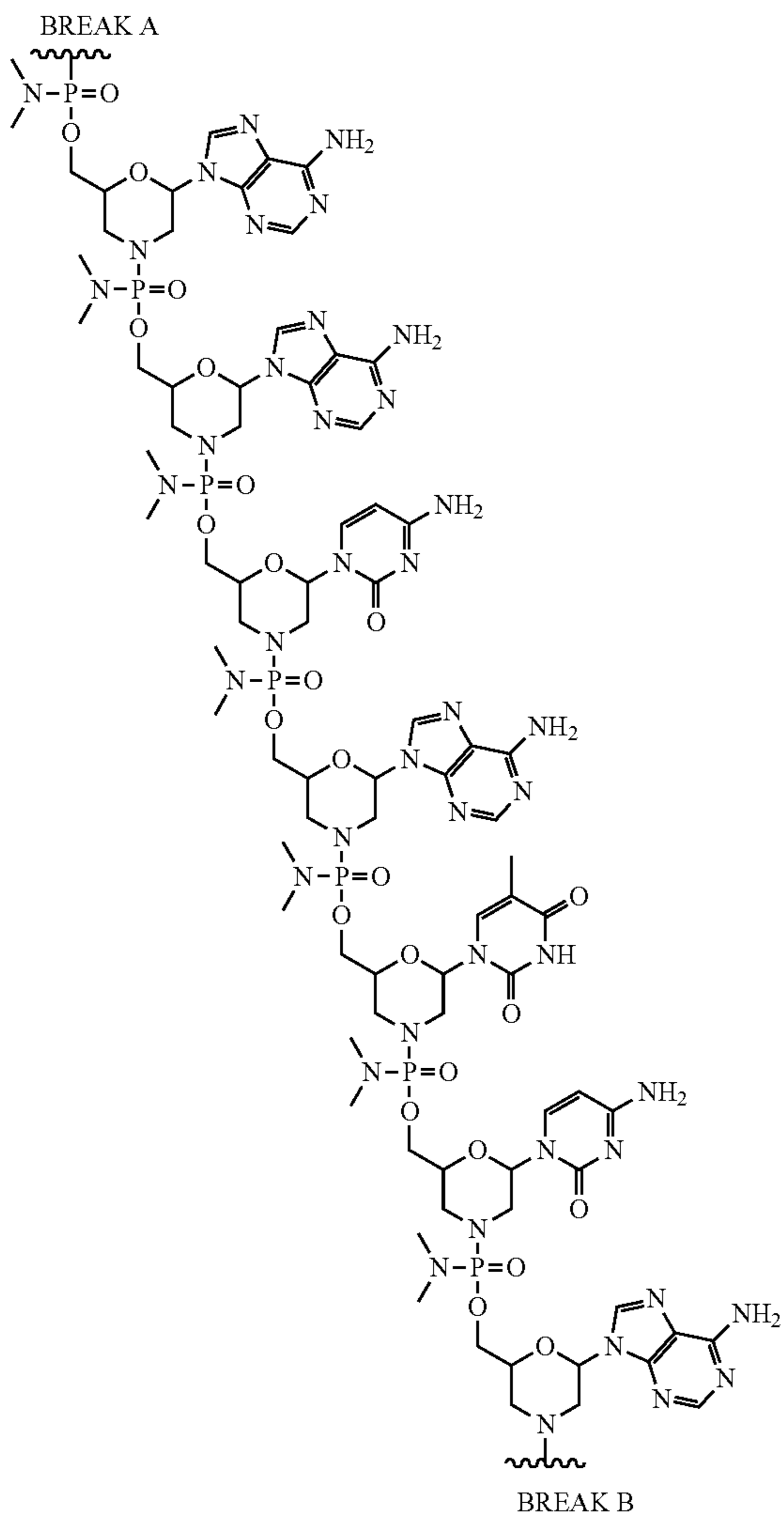
65

(E)



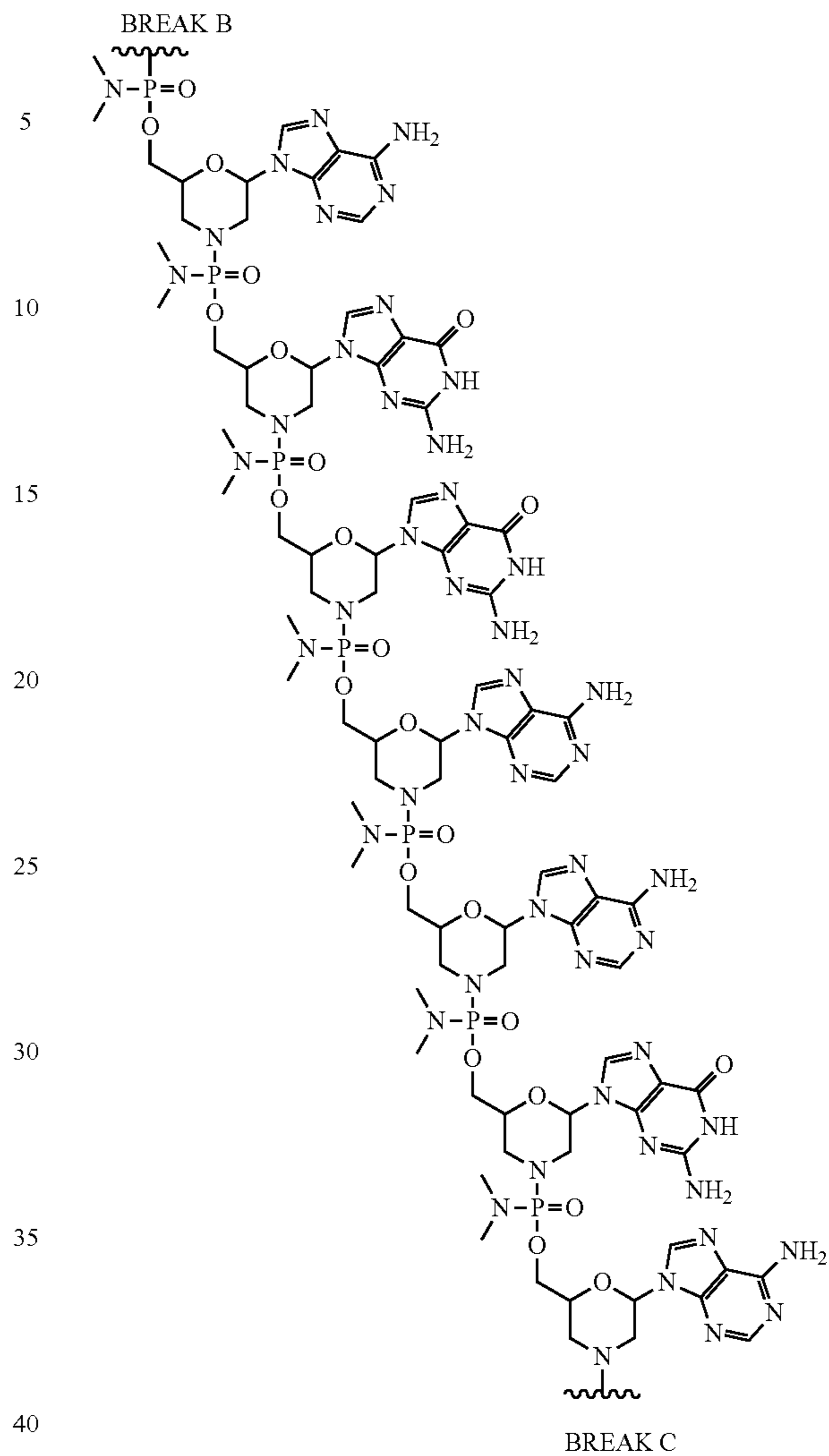
115

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116

-continued



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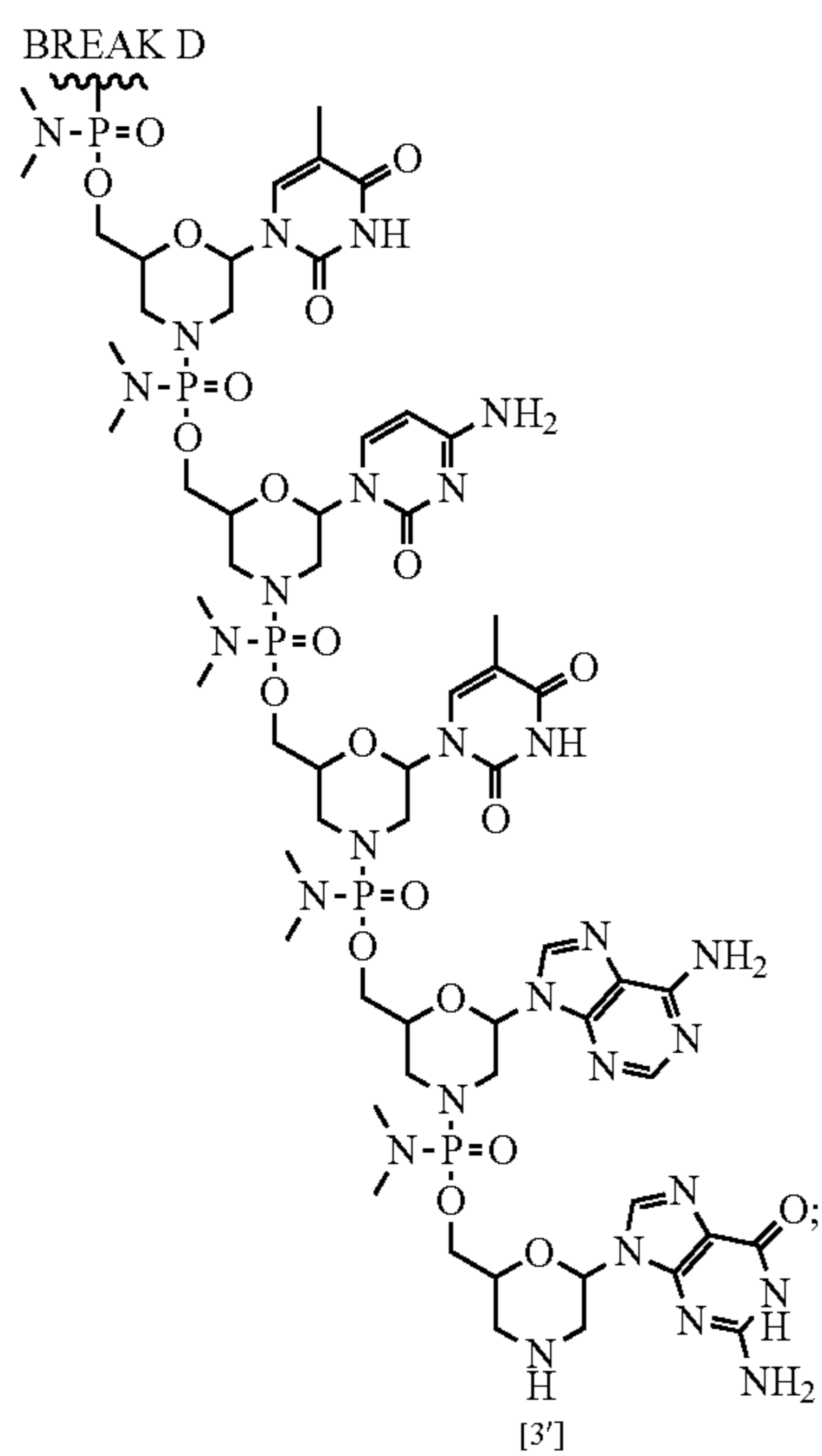
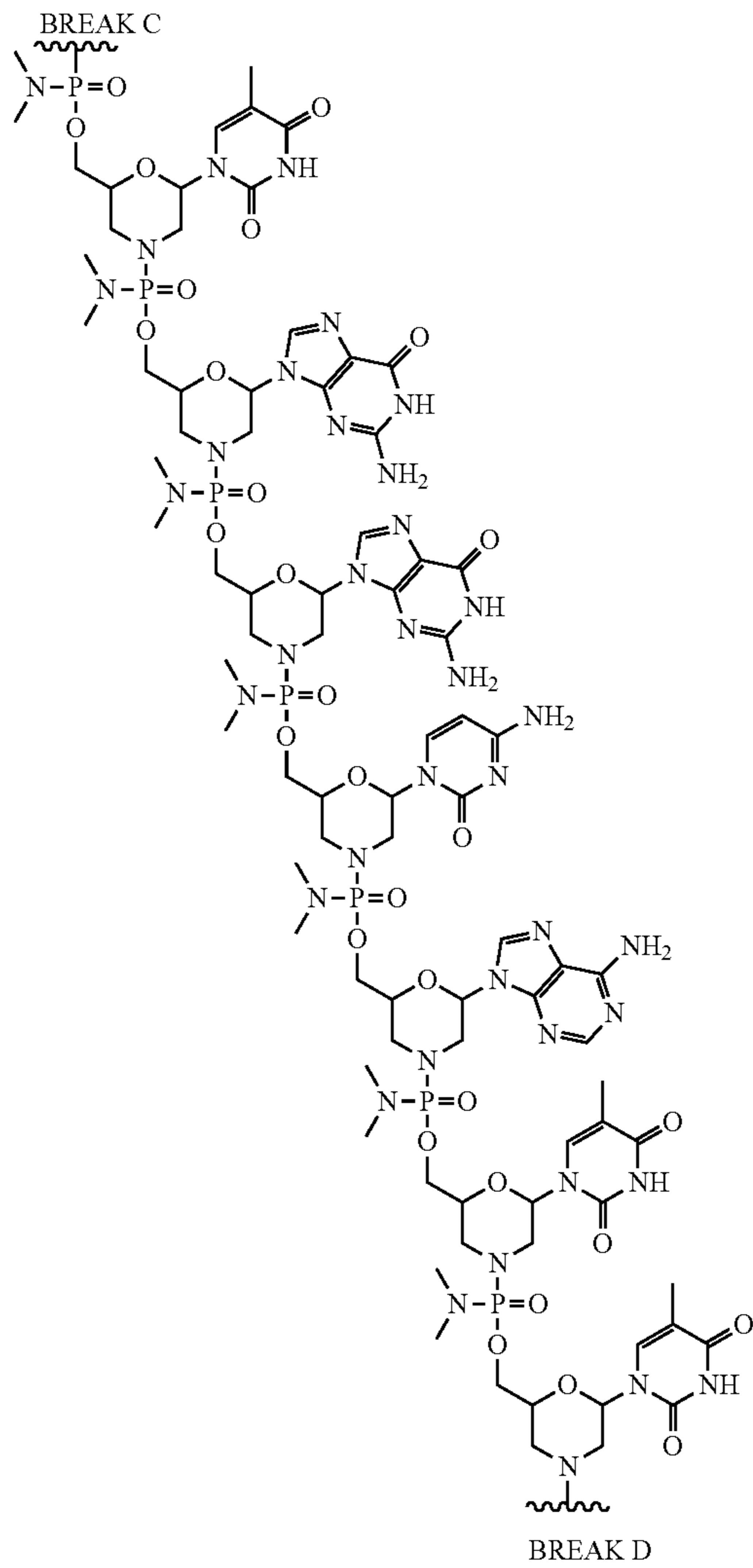
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117

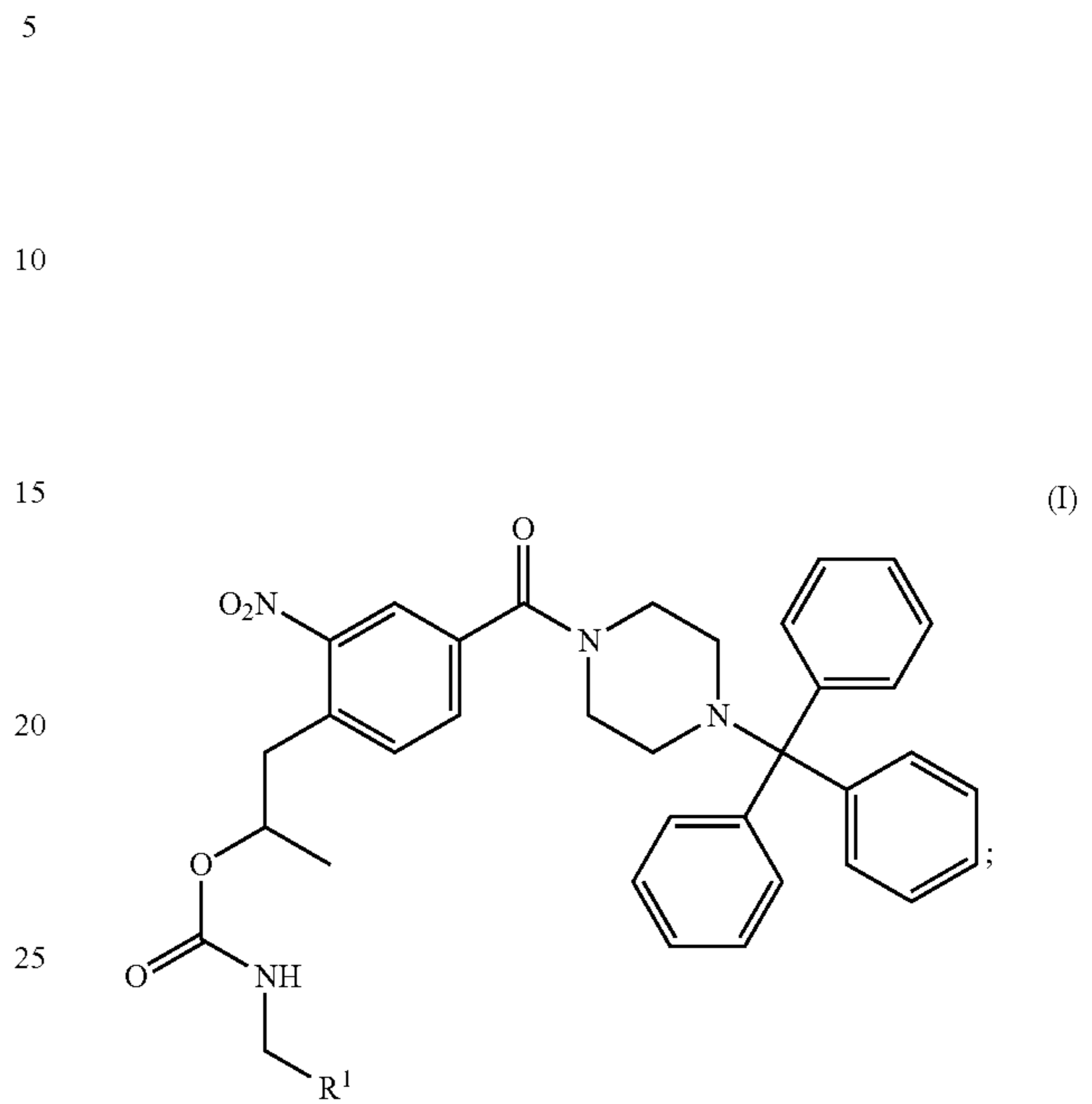
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118

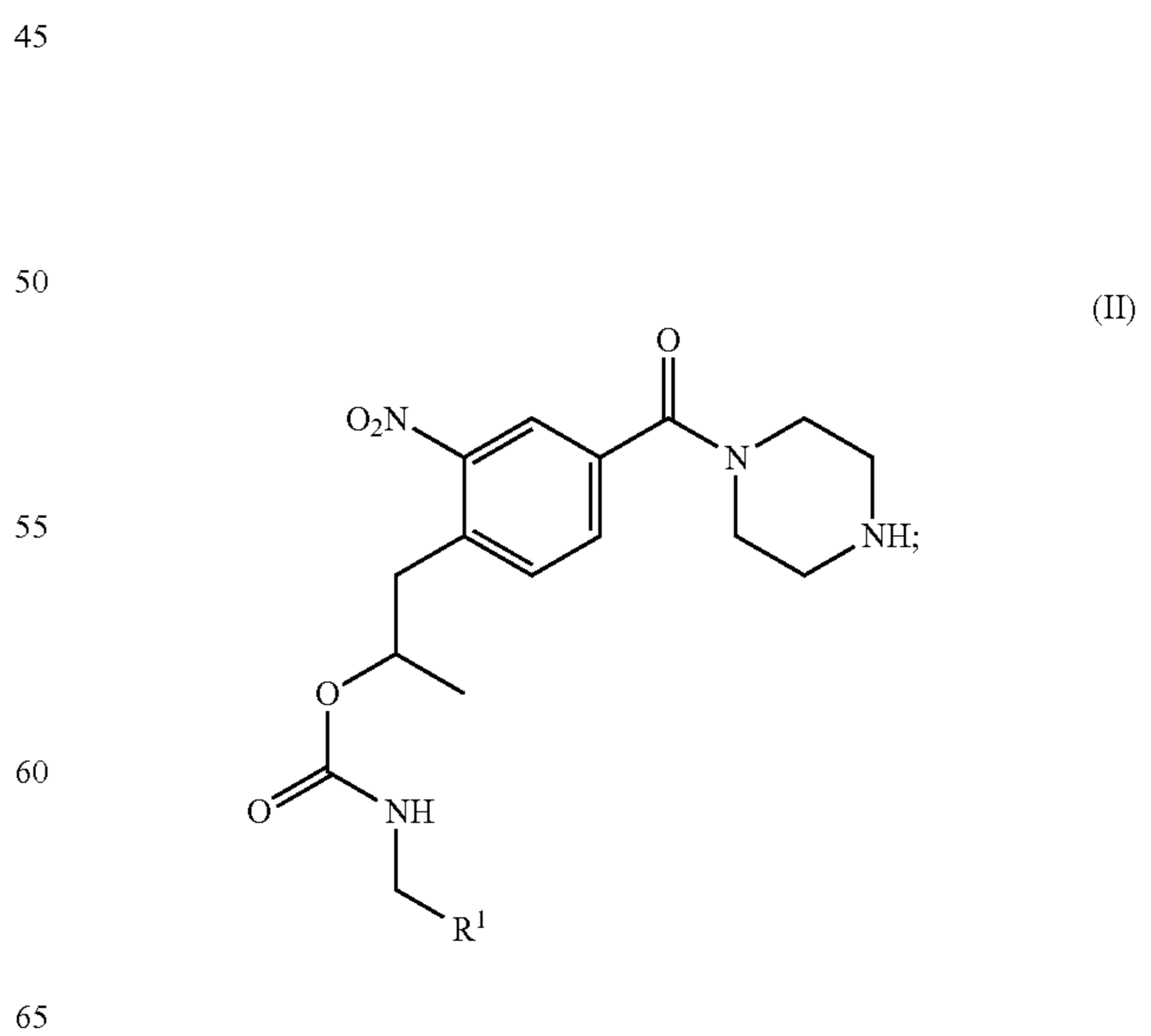
wherein the process comprises the sequential steps of:

(a) contacting a compound of Formula (I):



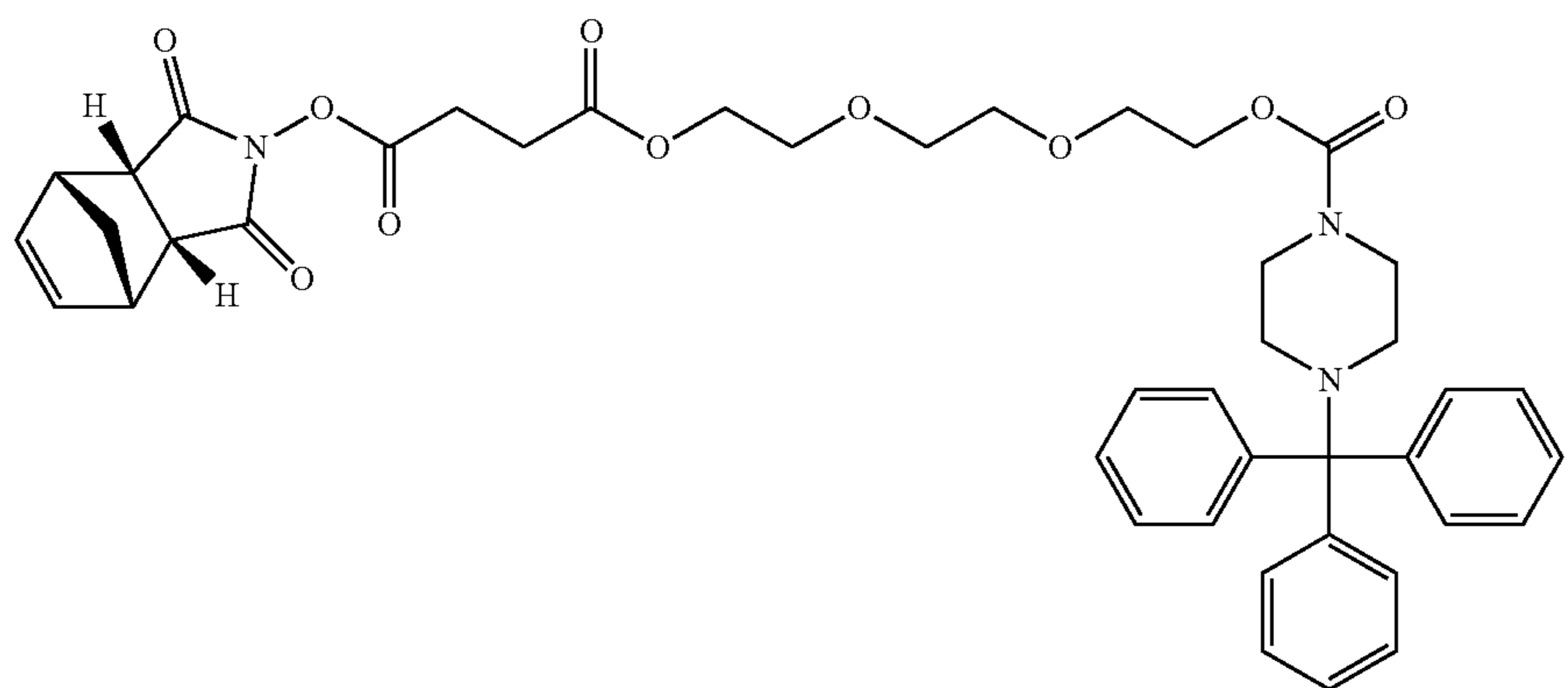
wherein R¹ is a support-medium,

with a deblocking agent to form the compound of Formula (II):

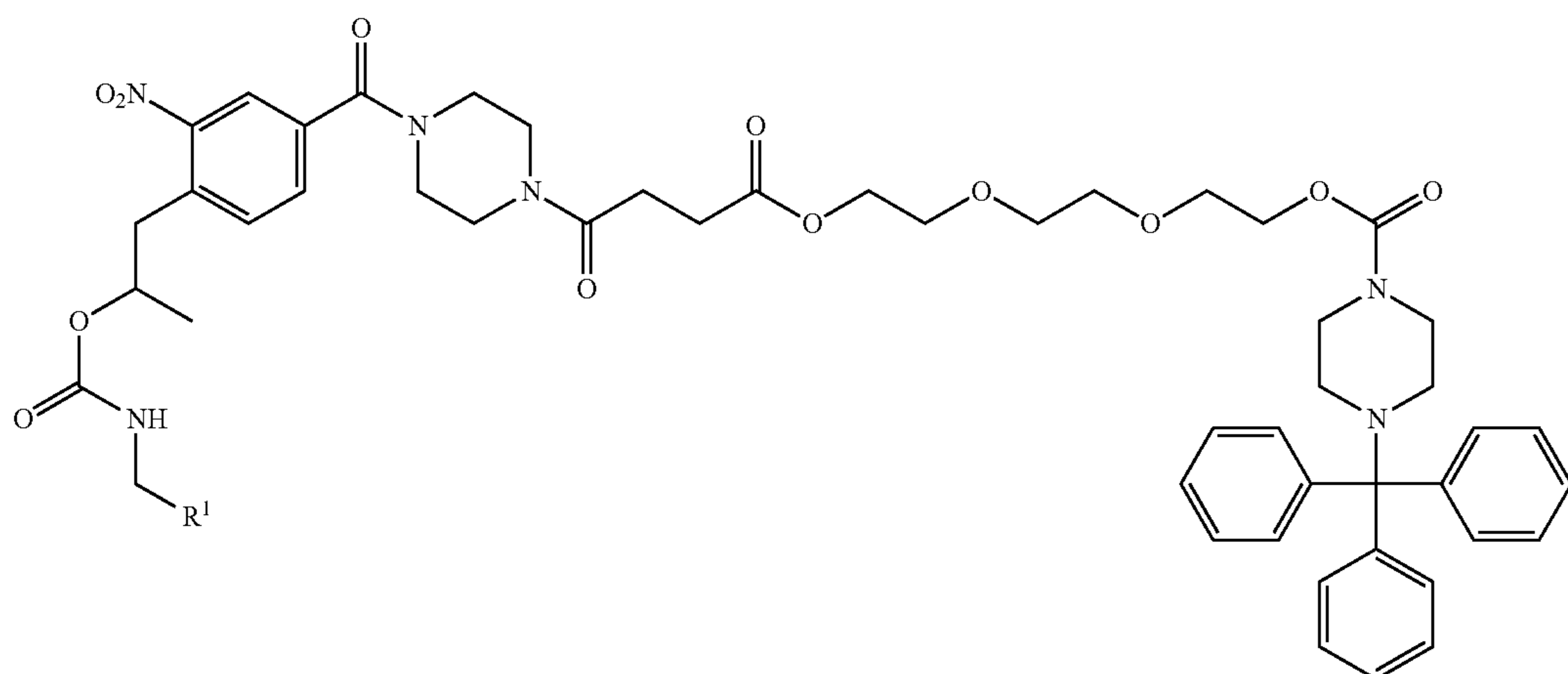


119

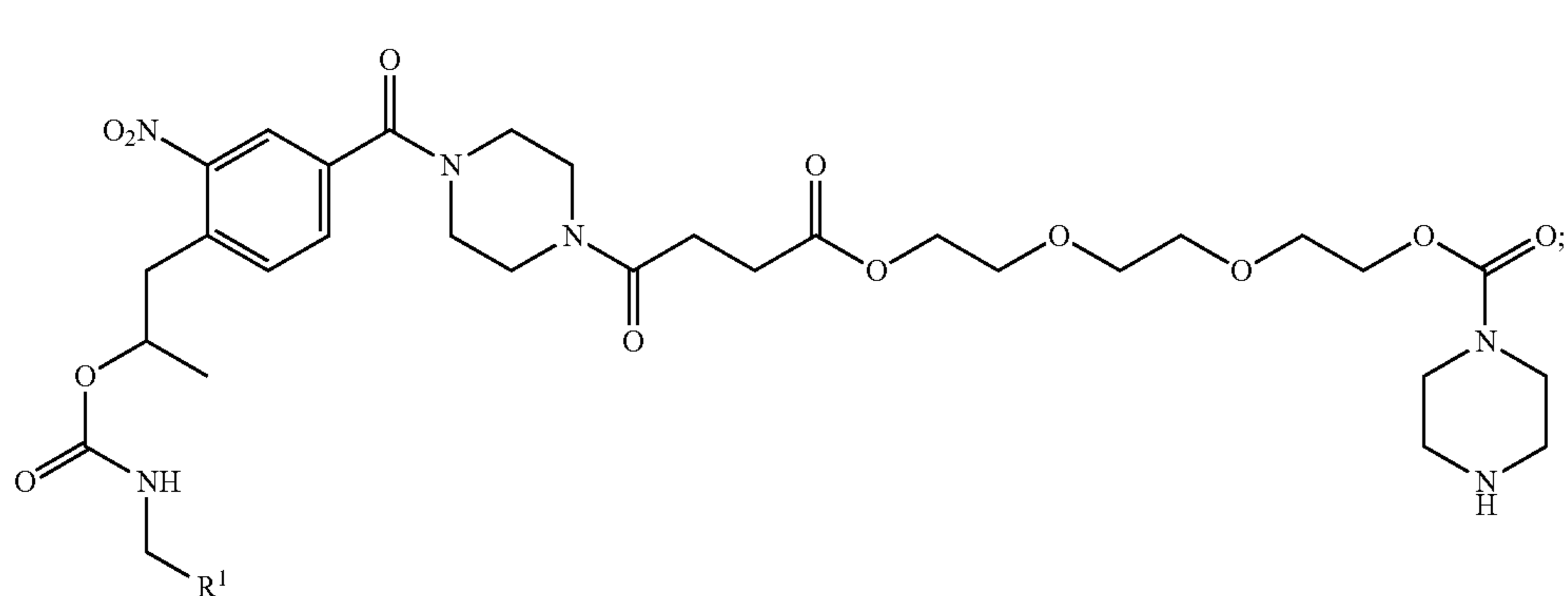
wherein R¹ is a support-medium;
 (b) contacting the compound of Formula (II) with compound
 (B):



to form a compound of Formula (III):

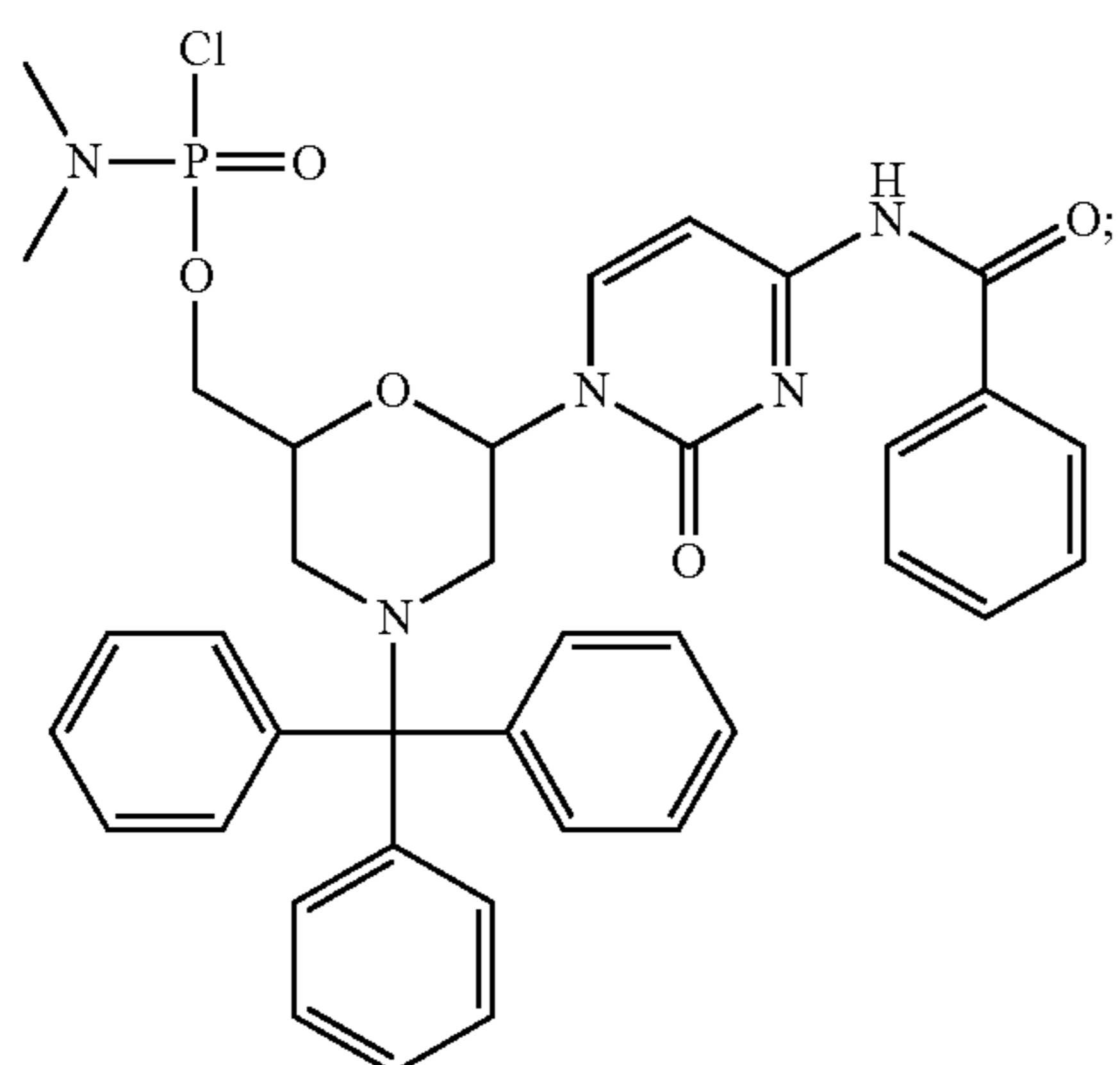


wherein R¹ is a support-medium;
 (c) contacting the compound of Formula (III) with a
 deblocking agent to form a compound of Formula (IV):



121

wherein R^1 is a support-medium;
 (d) contacting the compound of Formula (IV) with a compound of Formula (C):



(C)

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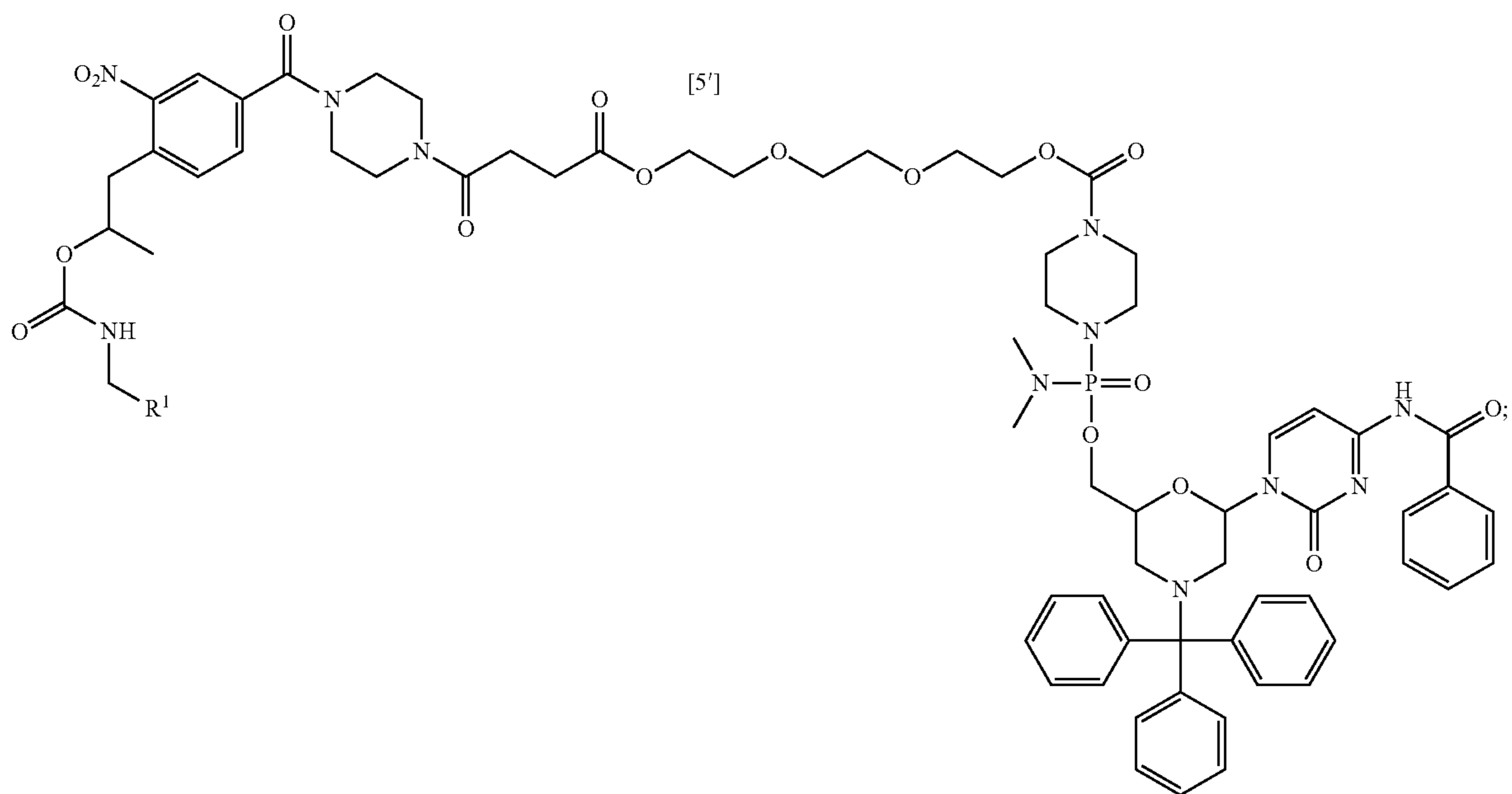
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to form a compound of Formula (V):



(V)

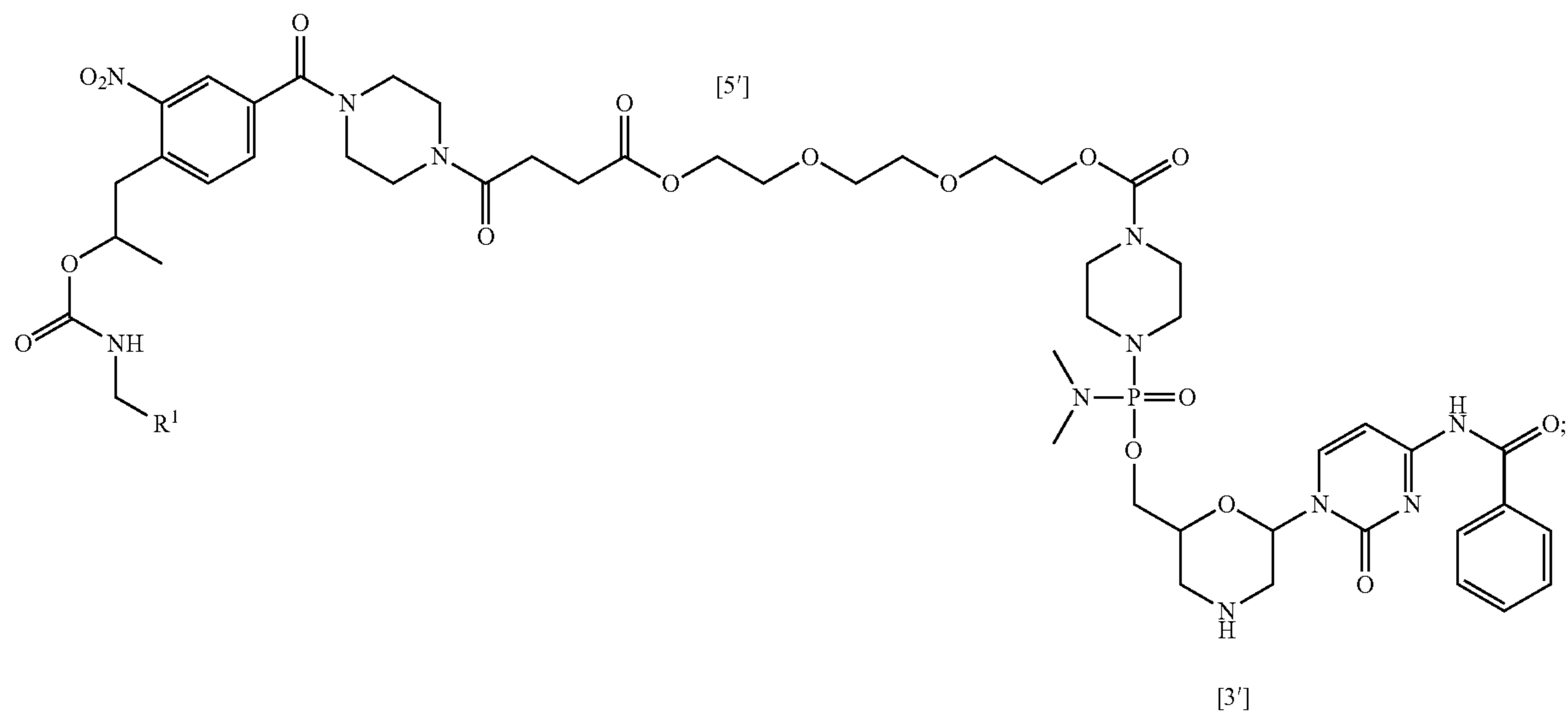
[3']

123

wherein R^1 is a support-medium;
 (e) contacting the compound of Formula (V) with a deblock-
 ing agent to form a compound of Formula (VI):

124

(VI)



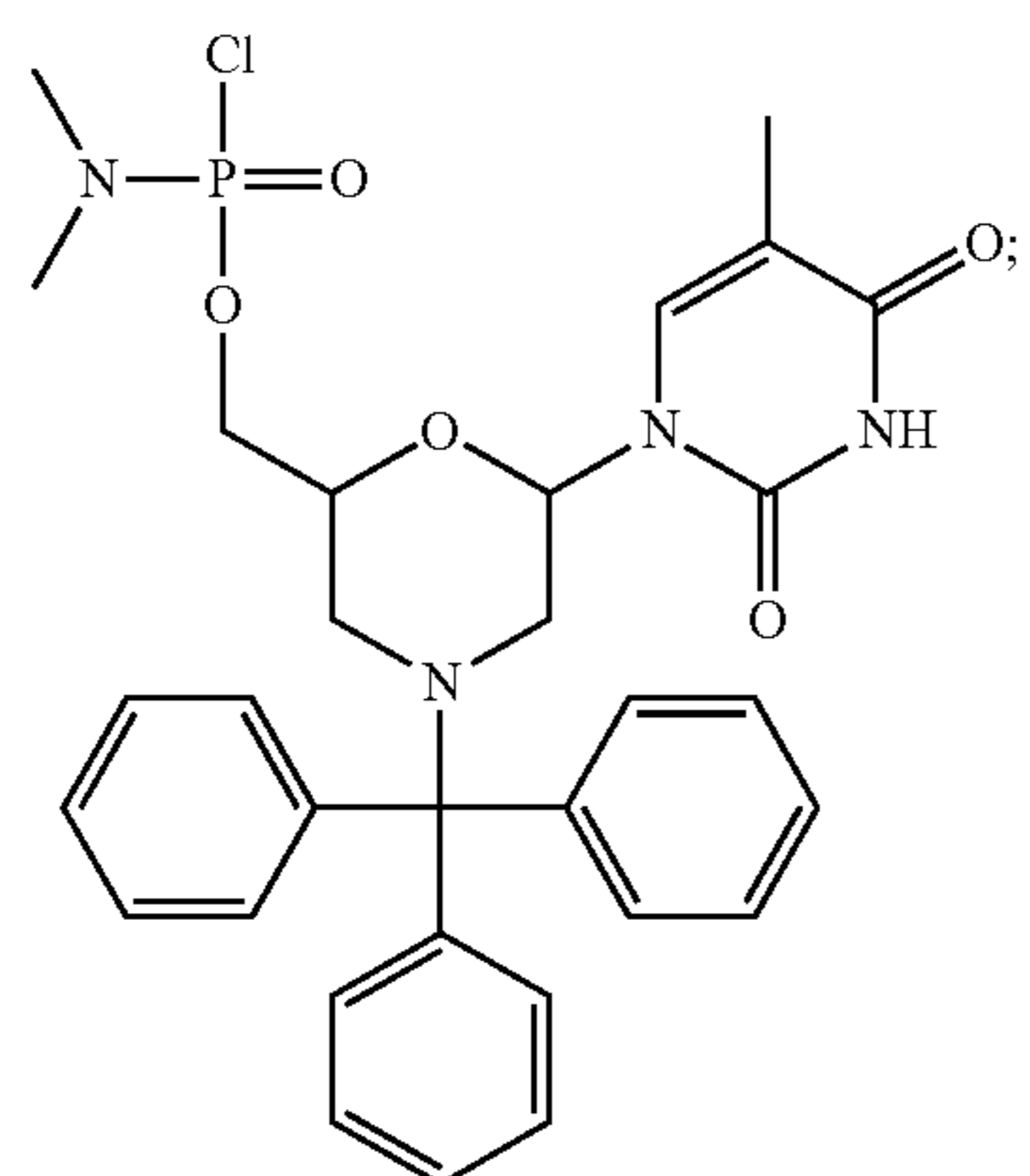
40

wherein R^1 is a support-medium;
 (f) contacting the compound of Formula (VI) with a com-
 pound of Formula (F):

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(F)

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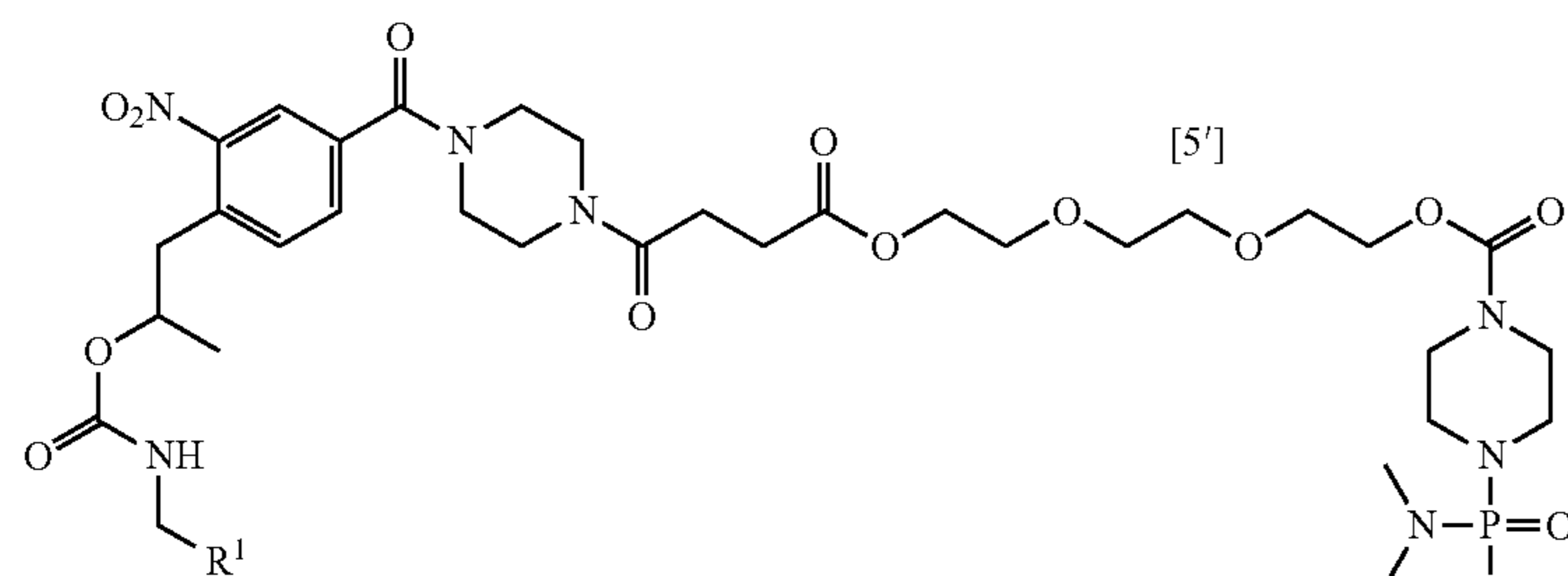
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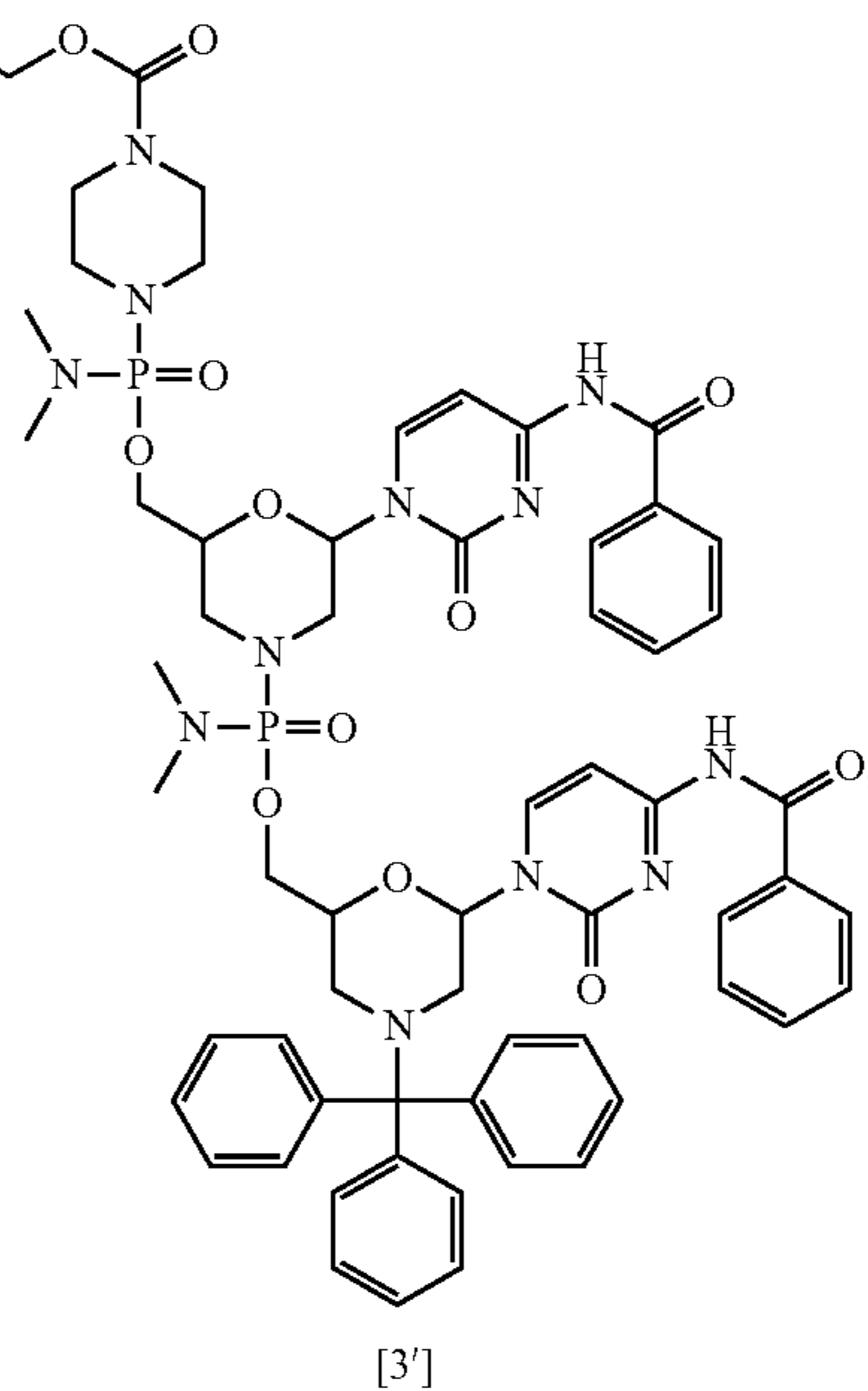
125

to form a compound of Formula (VII):



126

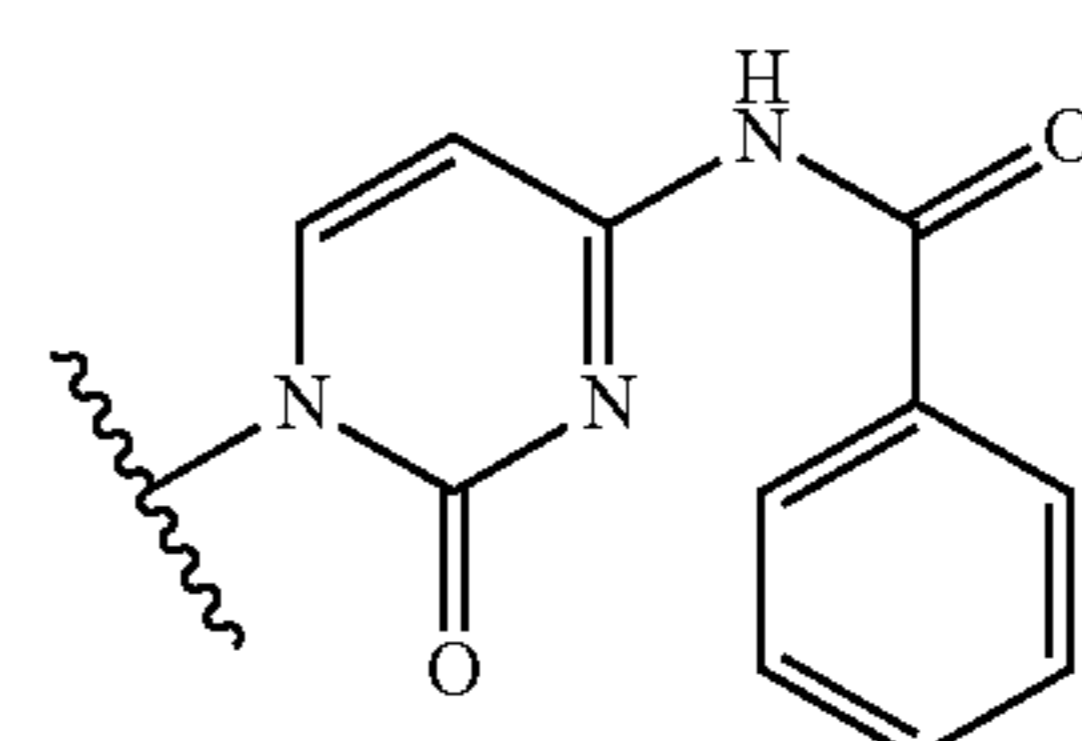
(VII)

wherein R¹ is a support-medium;

(g) performing 28 iterations of the sequential steps of:

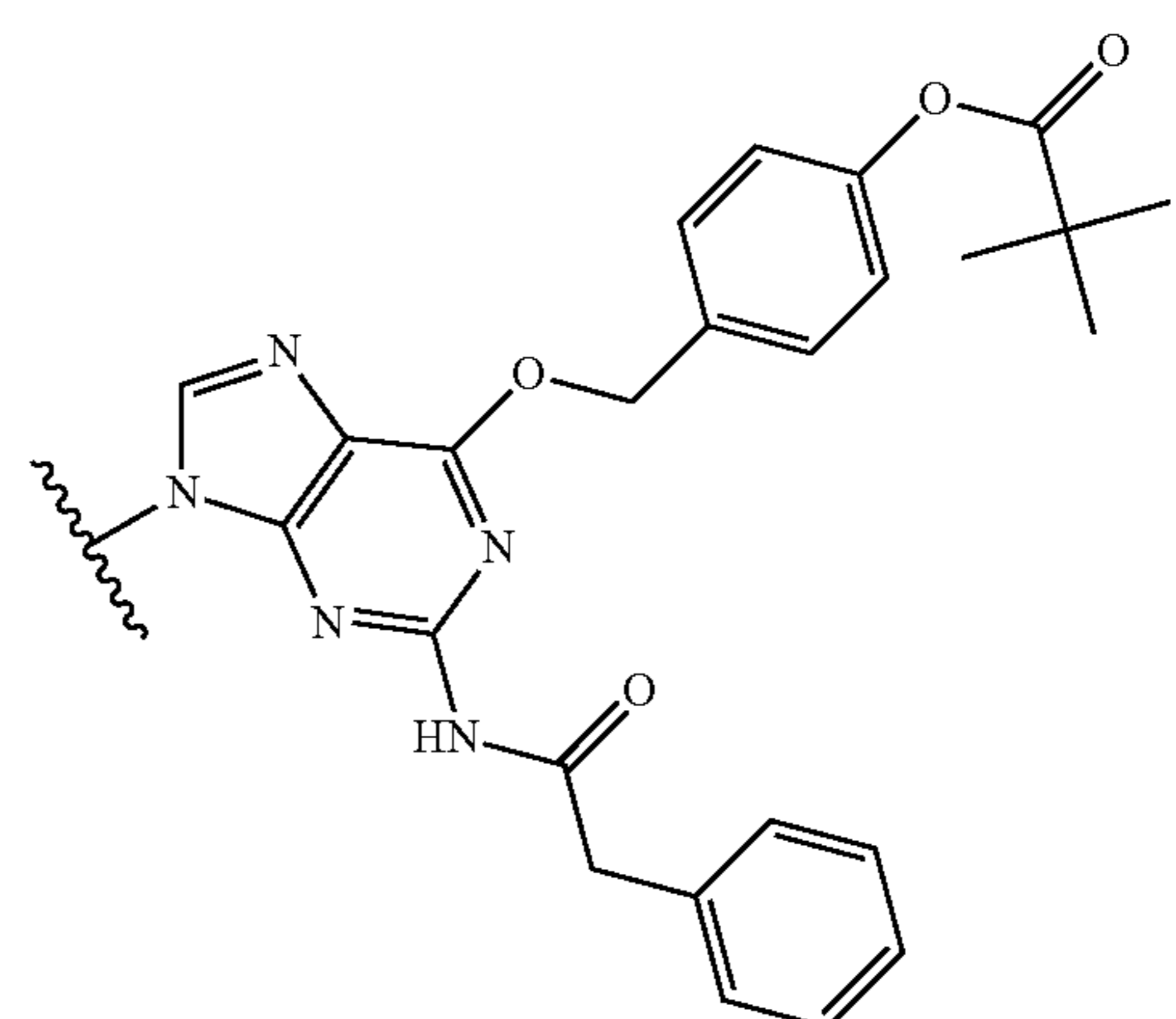
(g1) contacting the product formed by the immediately prior step with a deblocking agent; and

(g2) contacting the compound formed by the immediately prior step with a compound of Formula (VIII):



(PC)

40



(DPG)

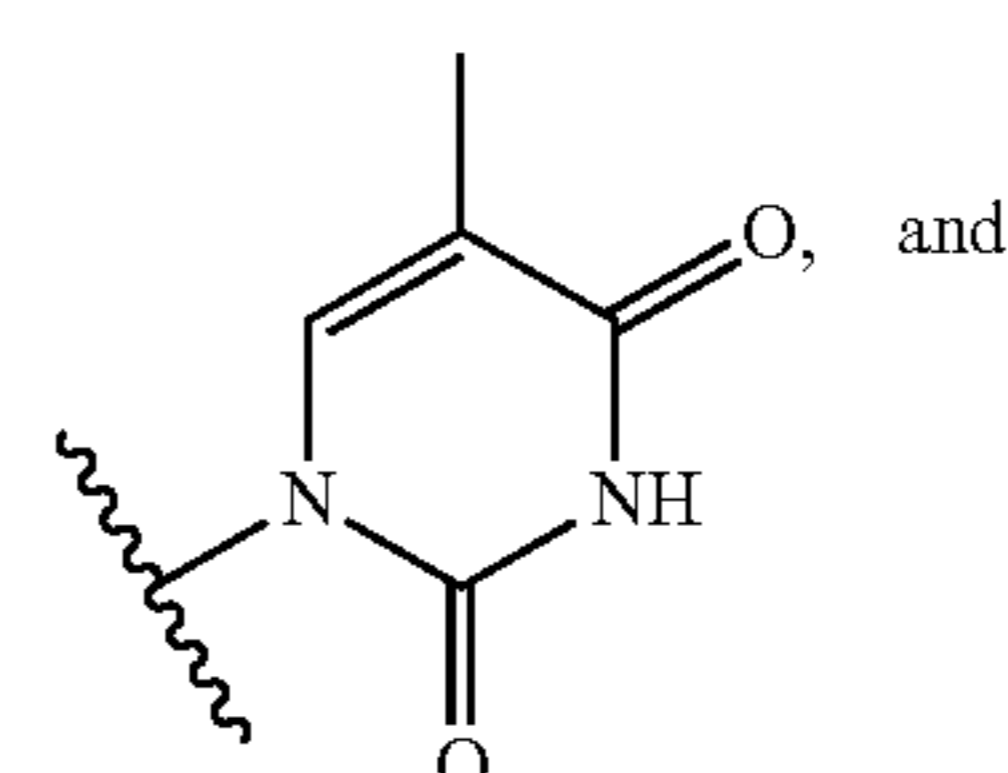
(VIII) 45

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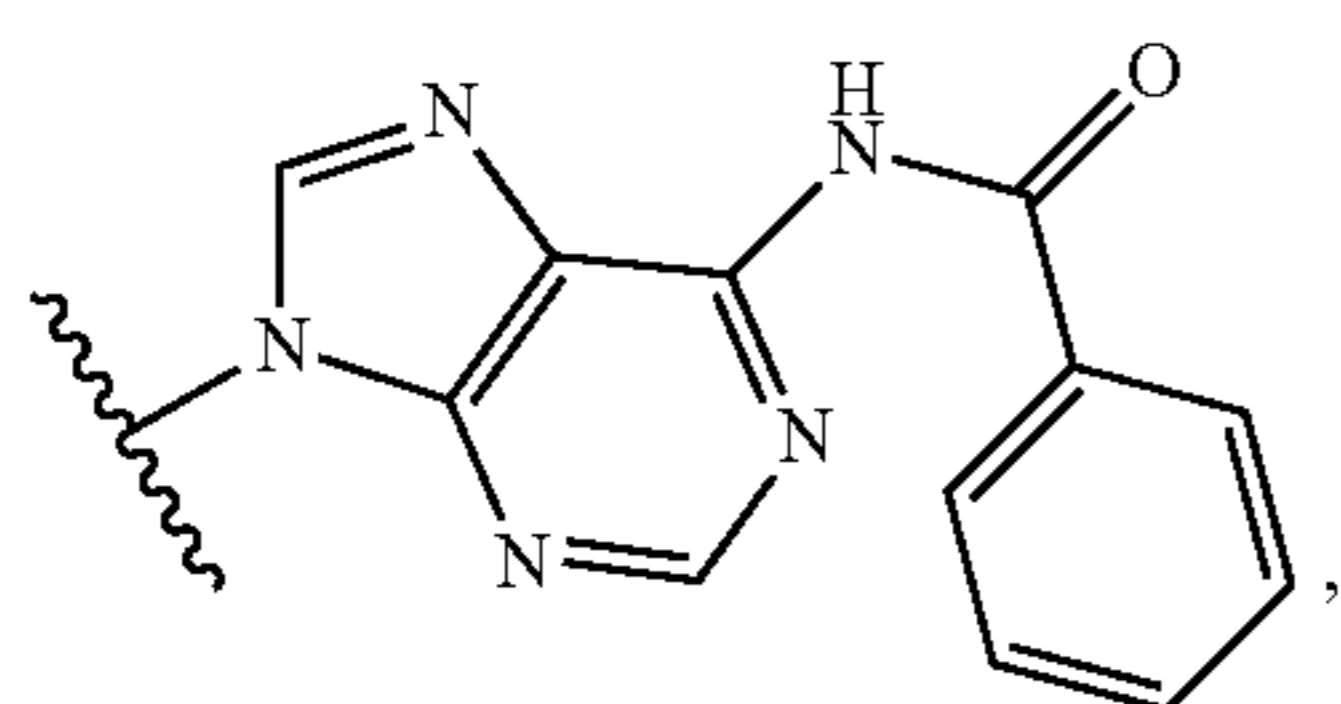
65

wherein R² is, independently for each compound of Formula (VIII), selected from the group consisting of:

(T)

127

-continued



wherein, for each iteration from 1 to 28, R² is:

Iteration No.	R ²
1	PC
2	PC
3	PA
4	PA
5	PC
6	PA

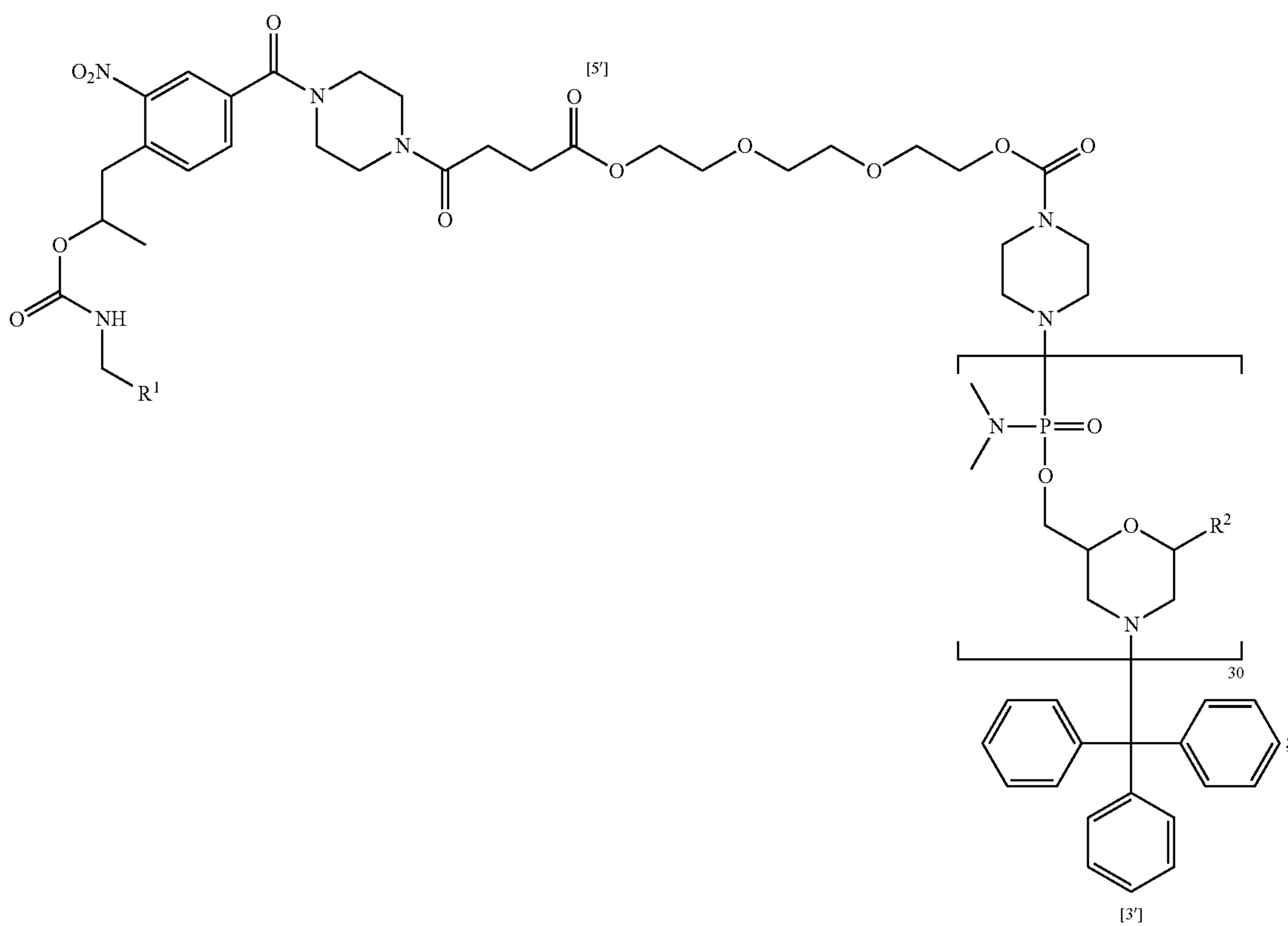
128

-continued

(PA)

	Iteration No.	R ²
5	7	T
	8	PC
	9	PA
10	10	PA
	11	DPG
	12	DPG
	13	PA
	14	PA
	15	DPG
	16	PA
15	17	T
	18	DPG
	19	DPG
	20	PC
	21	PA
	22	T
	23	T
20	24	T
	25	PC
	26	T
	27	PA
	28	DPG

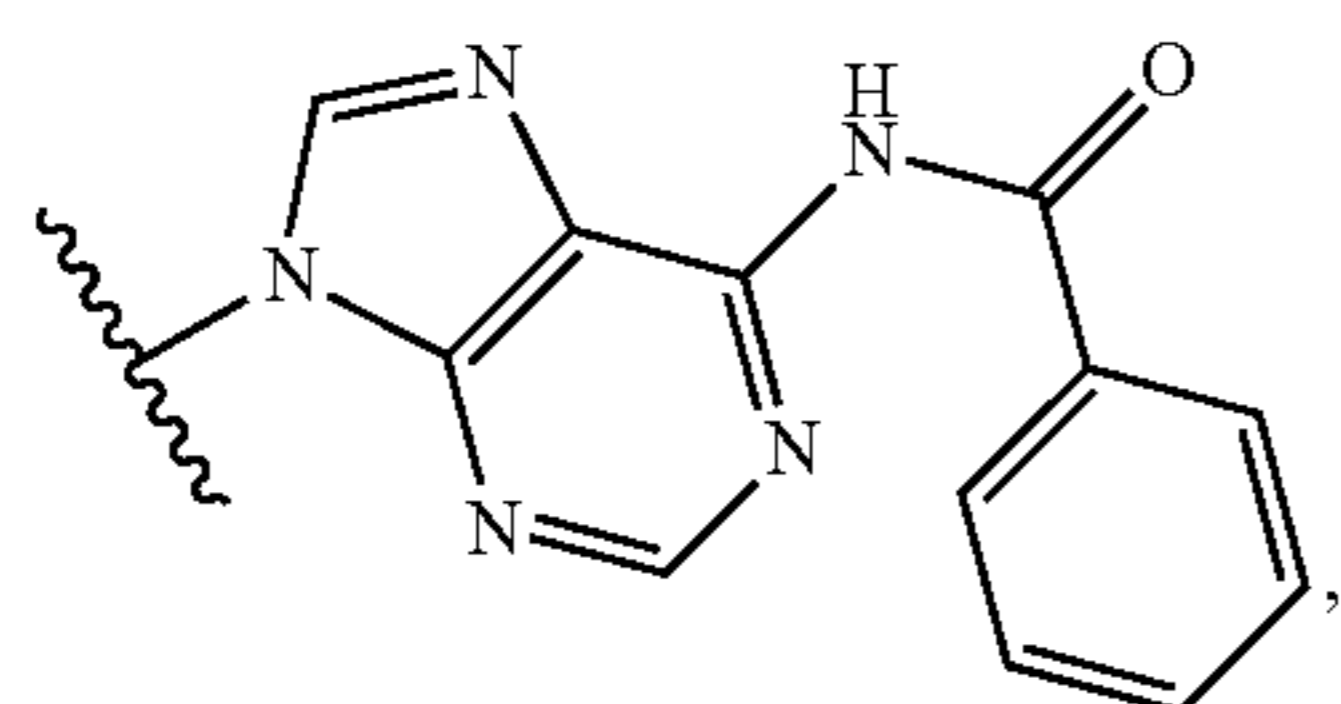
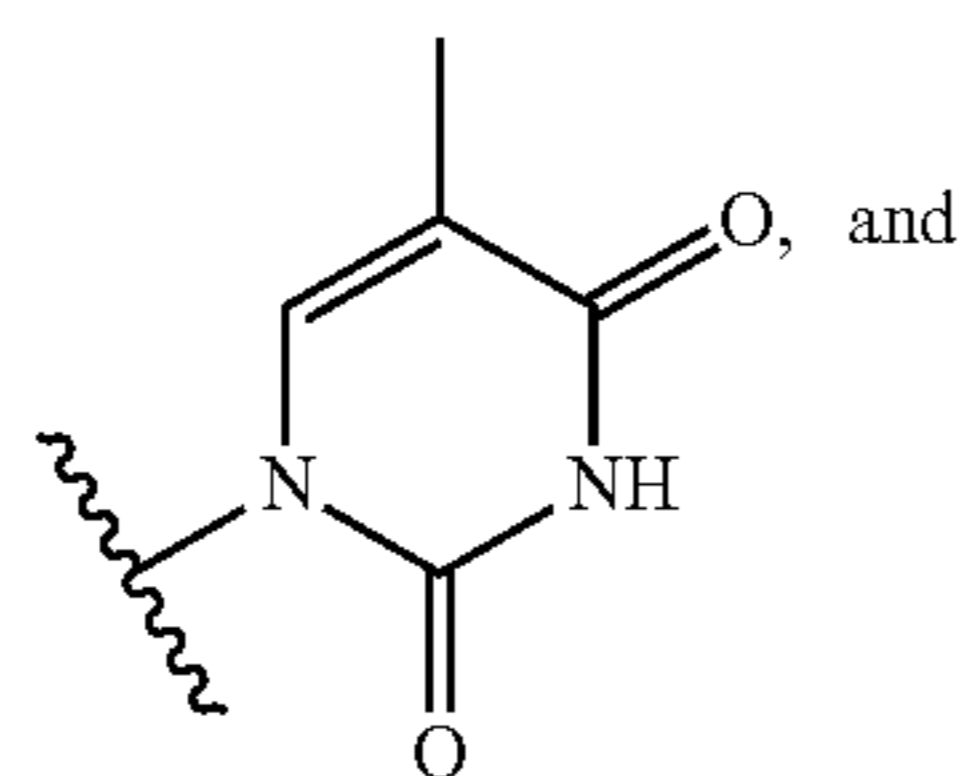
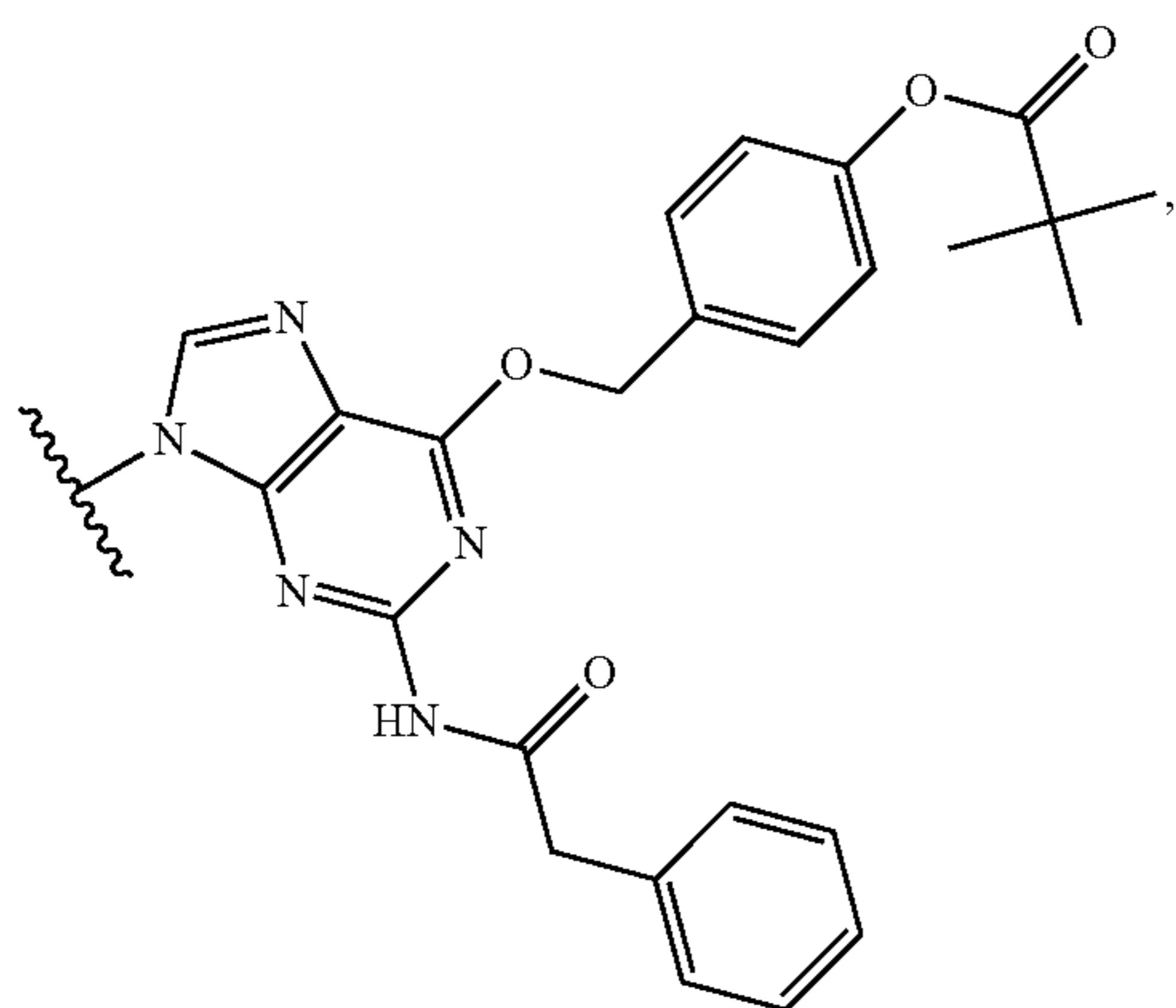
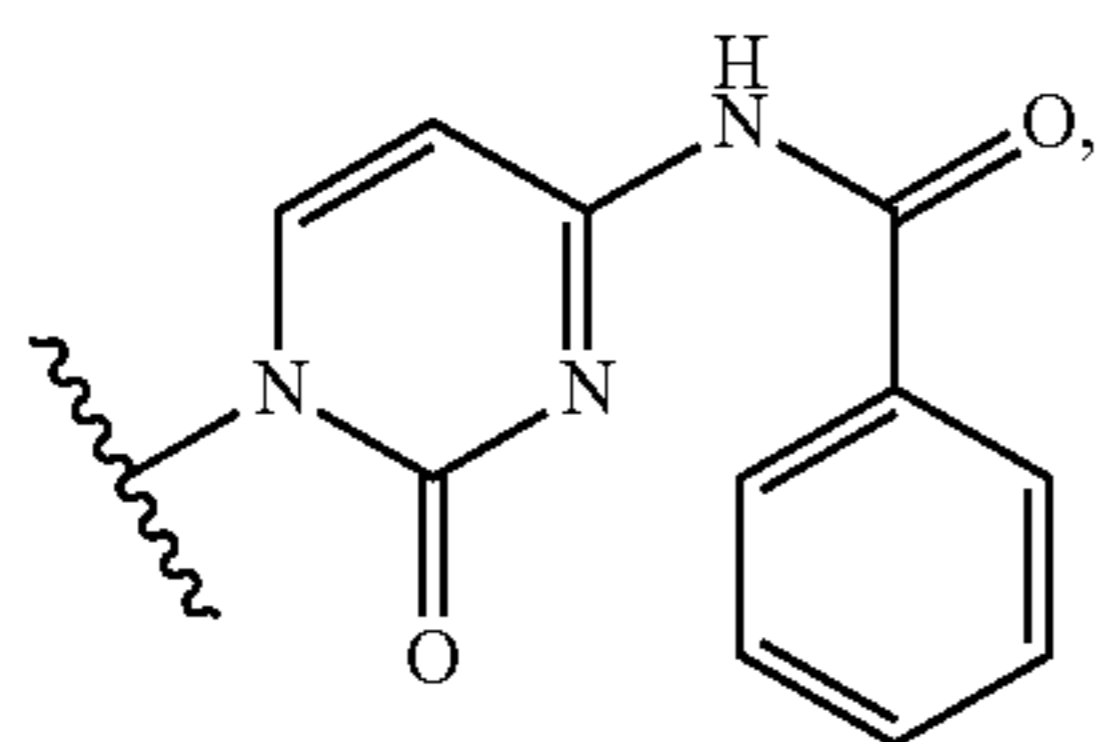
to form a compound of Formula (IX):



(IX)

129

wherein R¹ is a support-medium,
 wherein R² is, independently for each occurrence,
 selected from the group consisting of:



130

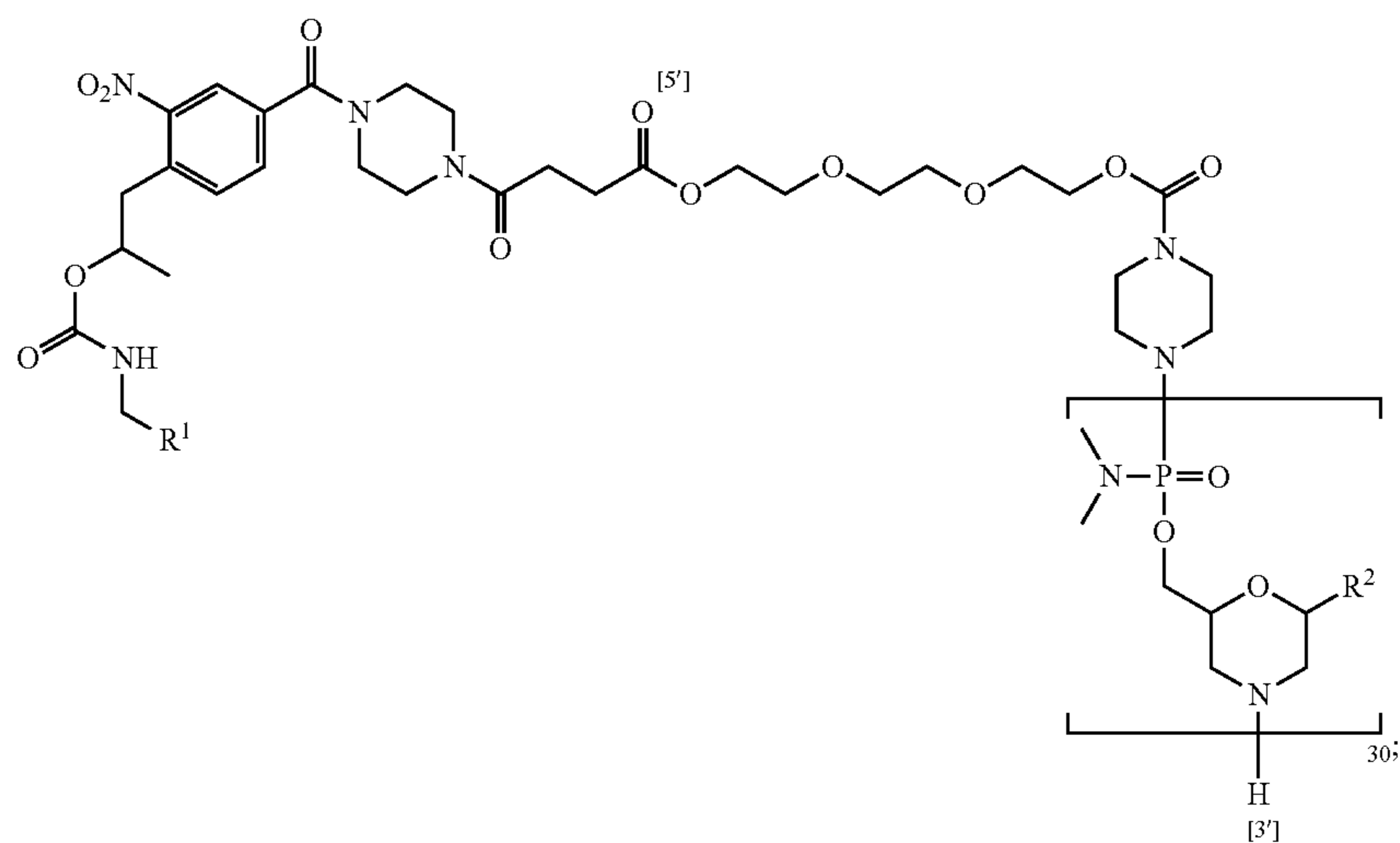
and

wherein R² is at each position from 1 to 30 and 5' to 3':

	Position No. 5' to 3'	R ²
(PC)	1	PC
	2	T
	3	PC
10	4	PC
	5	PA
	6	PA
(DPG)	7	PC
	8	PA
	9	T
15	10	PC
	11	PA
	12	PA
20	13	DPG
	14	DPG
	15	PA
25	16	PA
	17	DPG
	18	PA
(T)	19	T
	20	DPG
	21	DPG
30	22	PC
	23	PA
	24	T
35	25	T
	26	T
	27	PC
(PA)	28	T
	29	PA
	30	DPG

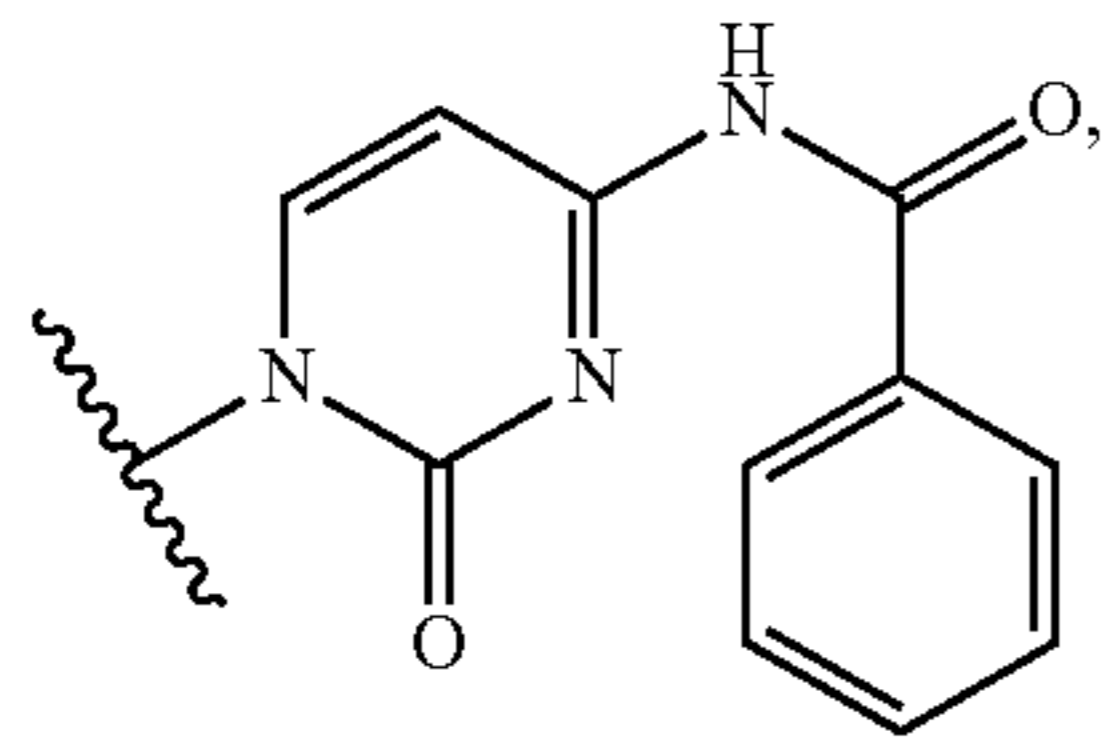
(h) contacting the compound of Formula (IX) with a deblocking agent to form a compound of Formula (X):

(X)



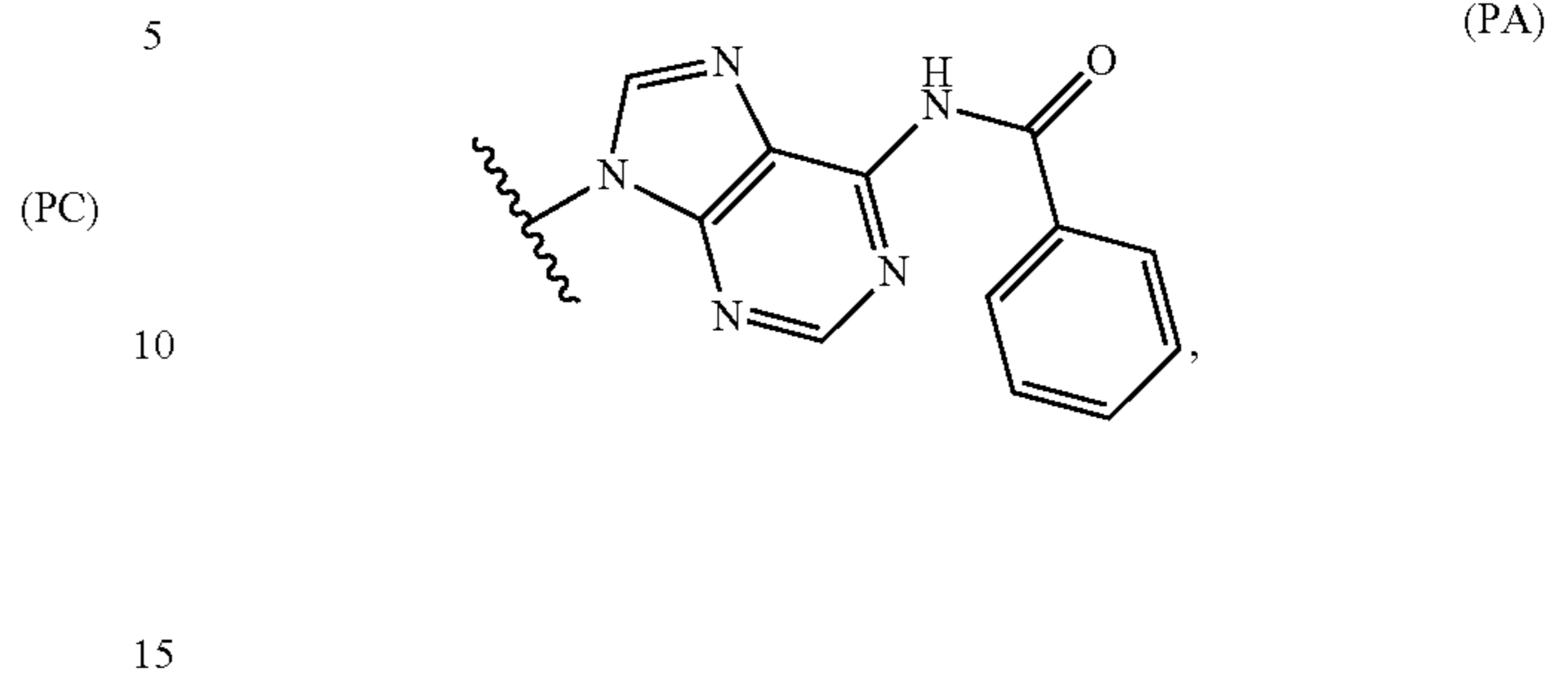
131

wherein R¹ is a support-medium,
 wherein R² is, independently for each occurrence,
 selected from the group consisting of:



132

-continued



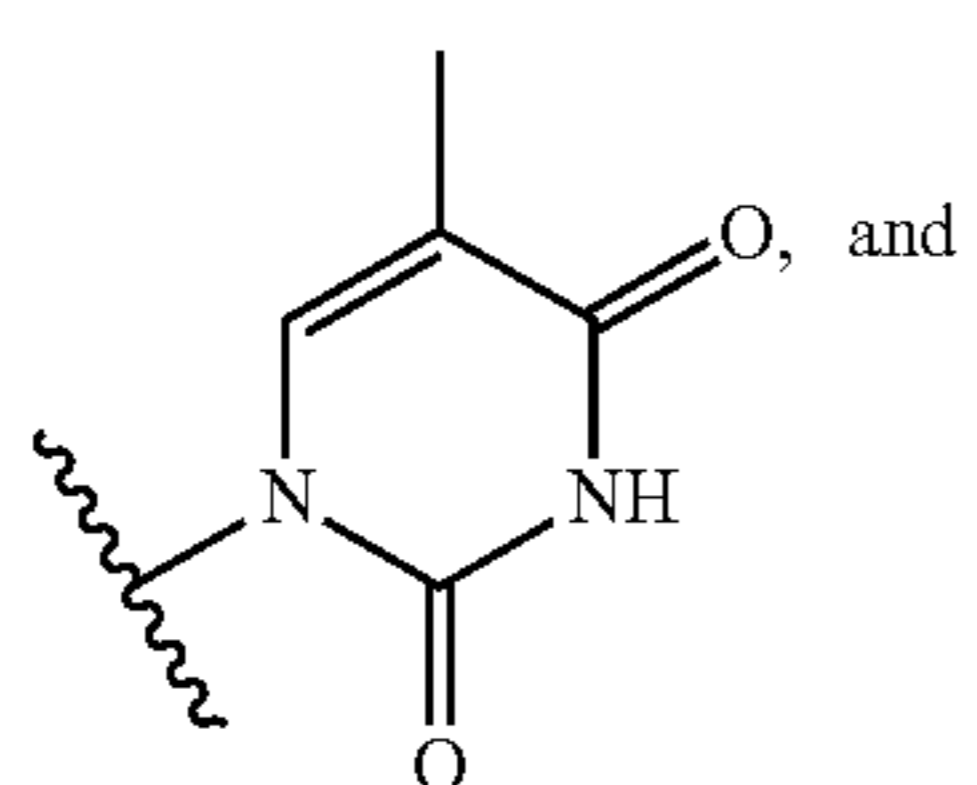
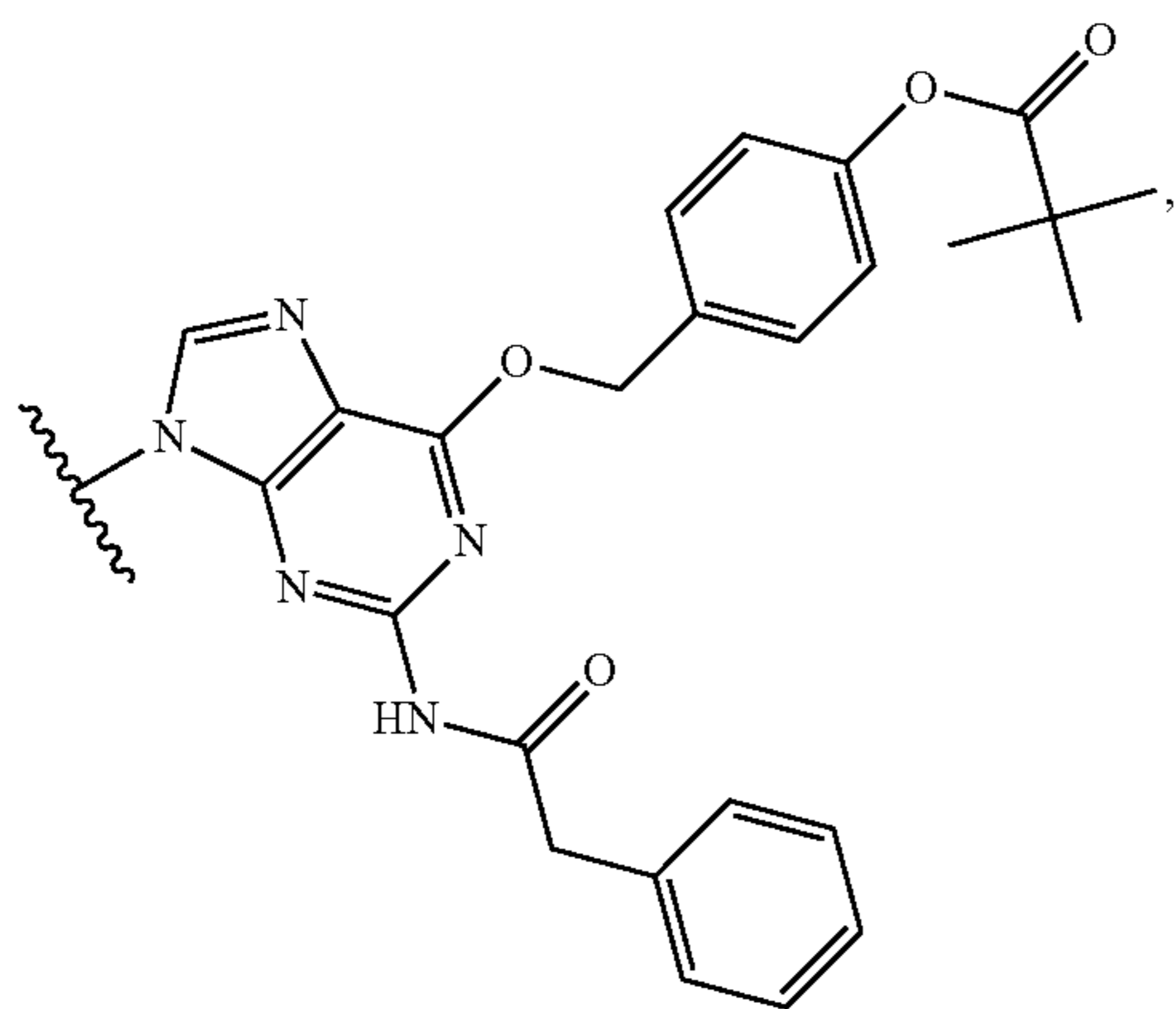
and

20 wherein R² is at each position from 1 to 30 and 5' to 3':

25

Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC
8	PA
9	T
10	PC
11	PA
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG
18	PA
19	T
20	DPG
21	DPG
22	PC
23	PA
24	T
25	T
26	T
27	PC
28	T
29	PA
30	DPG

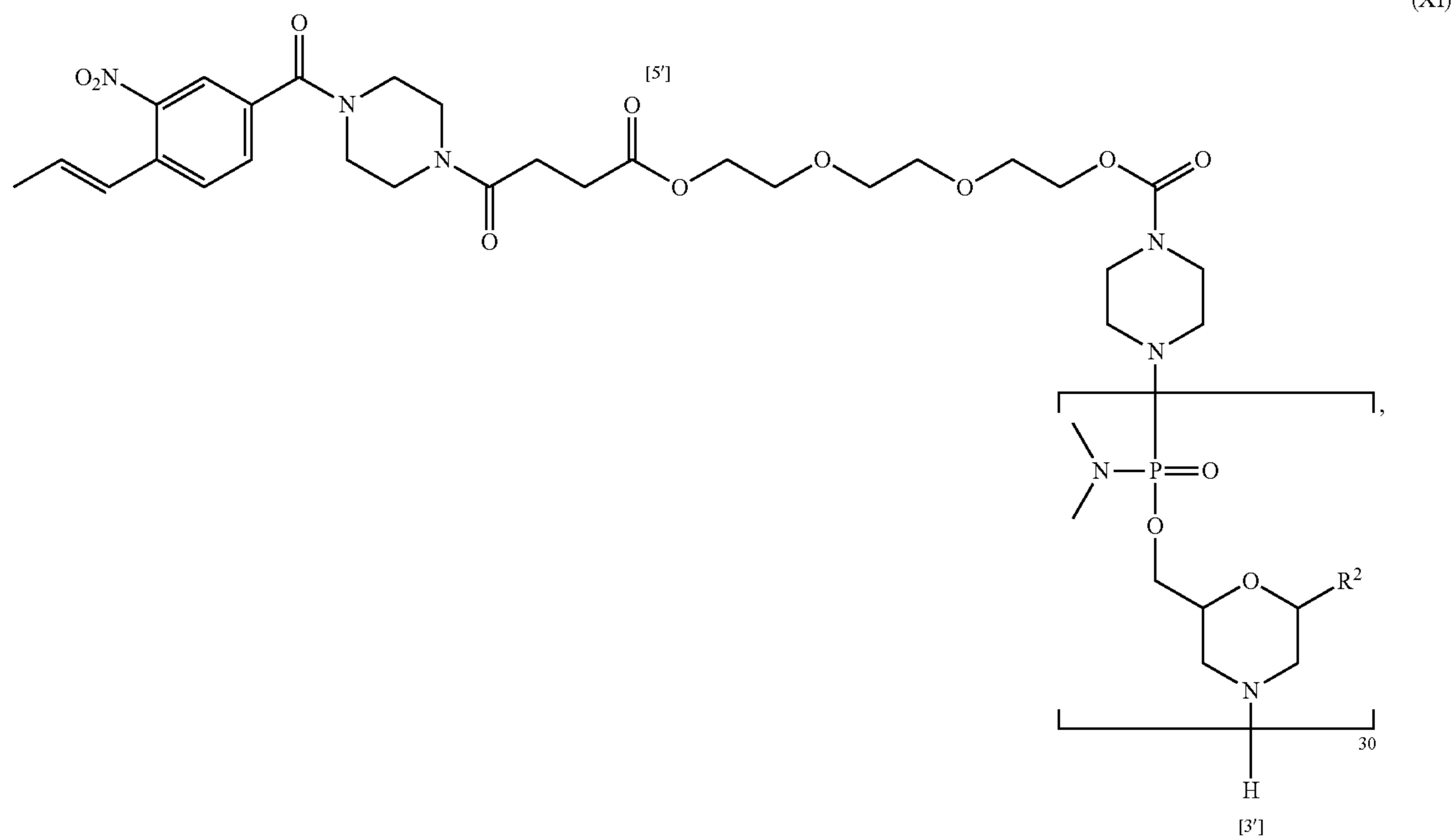
65



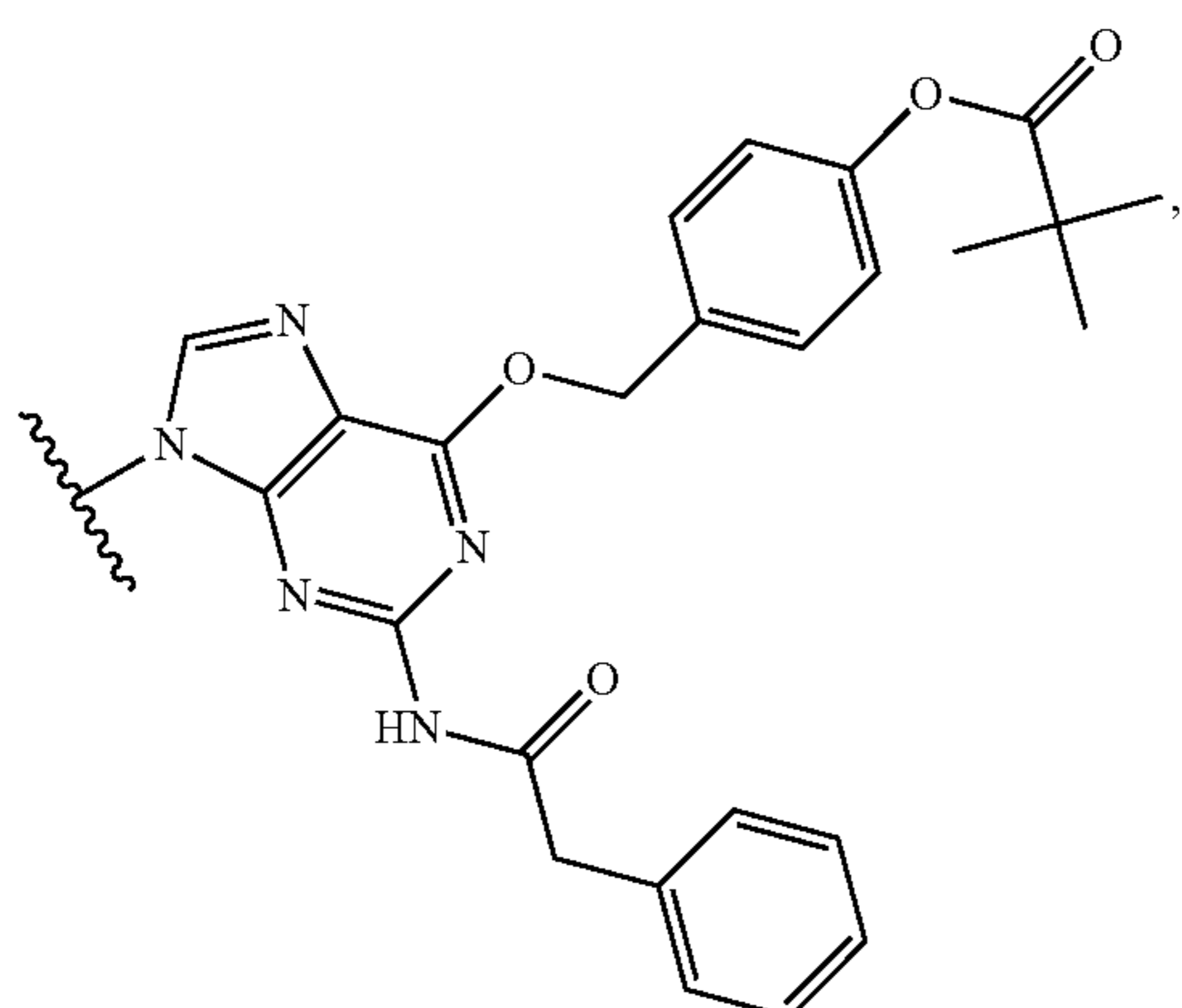
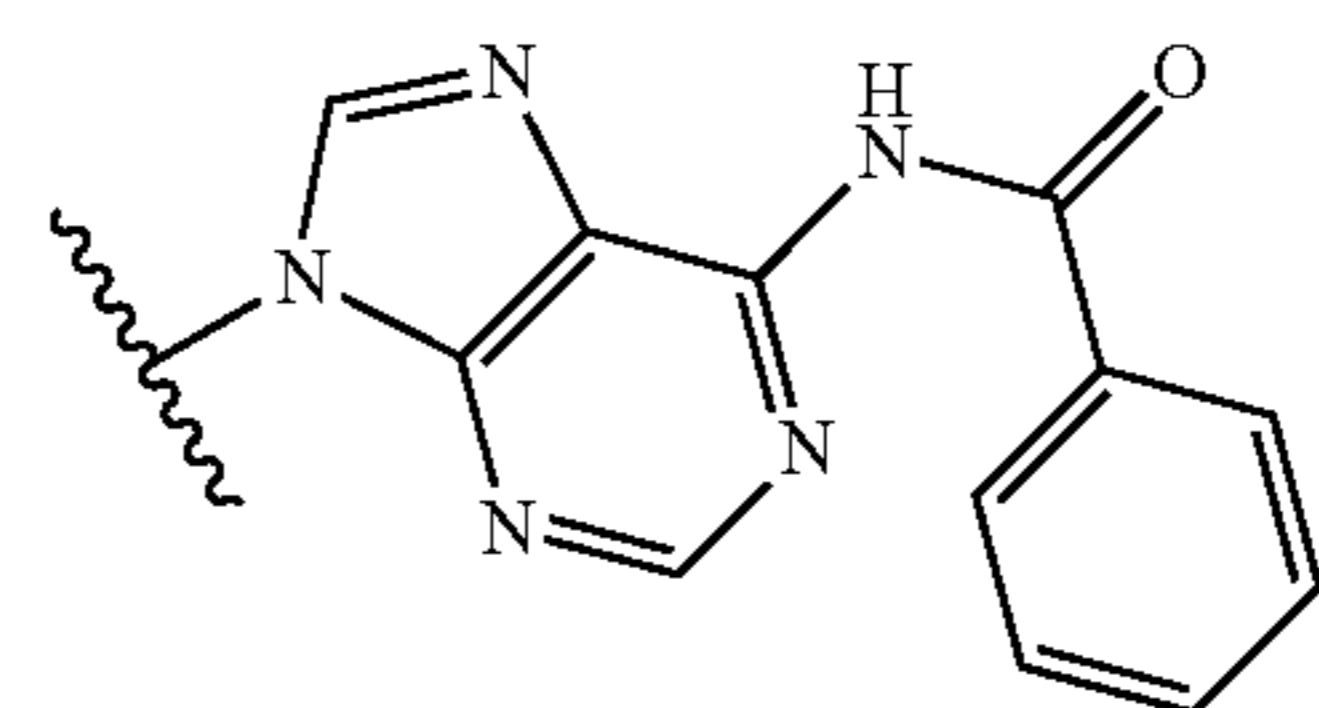
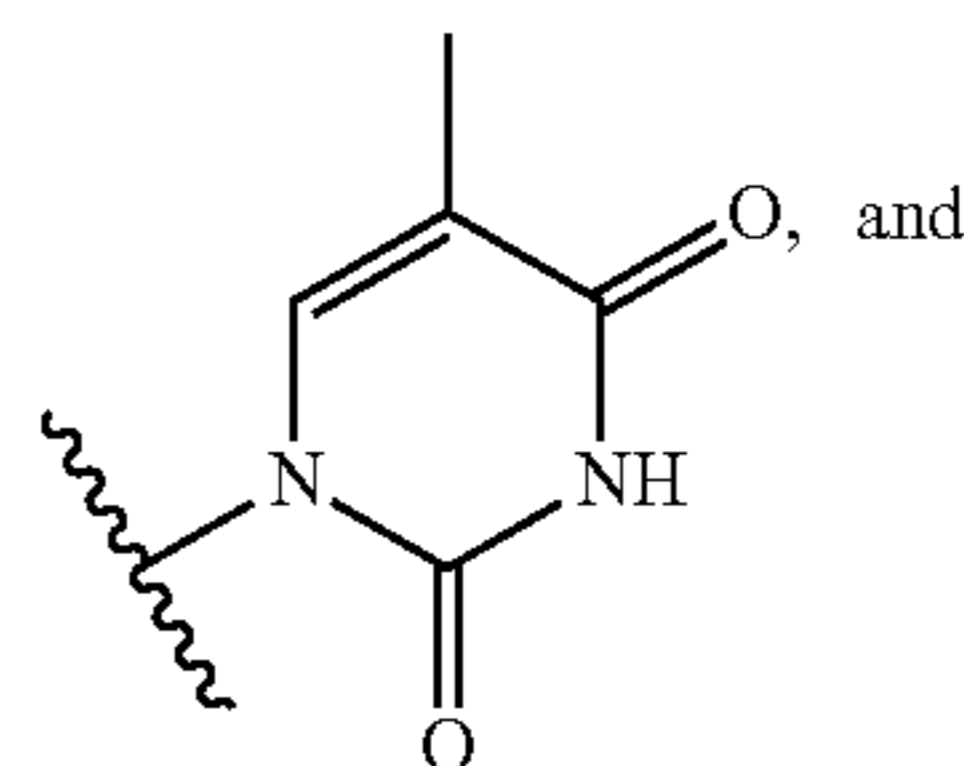
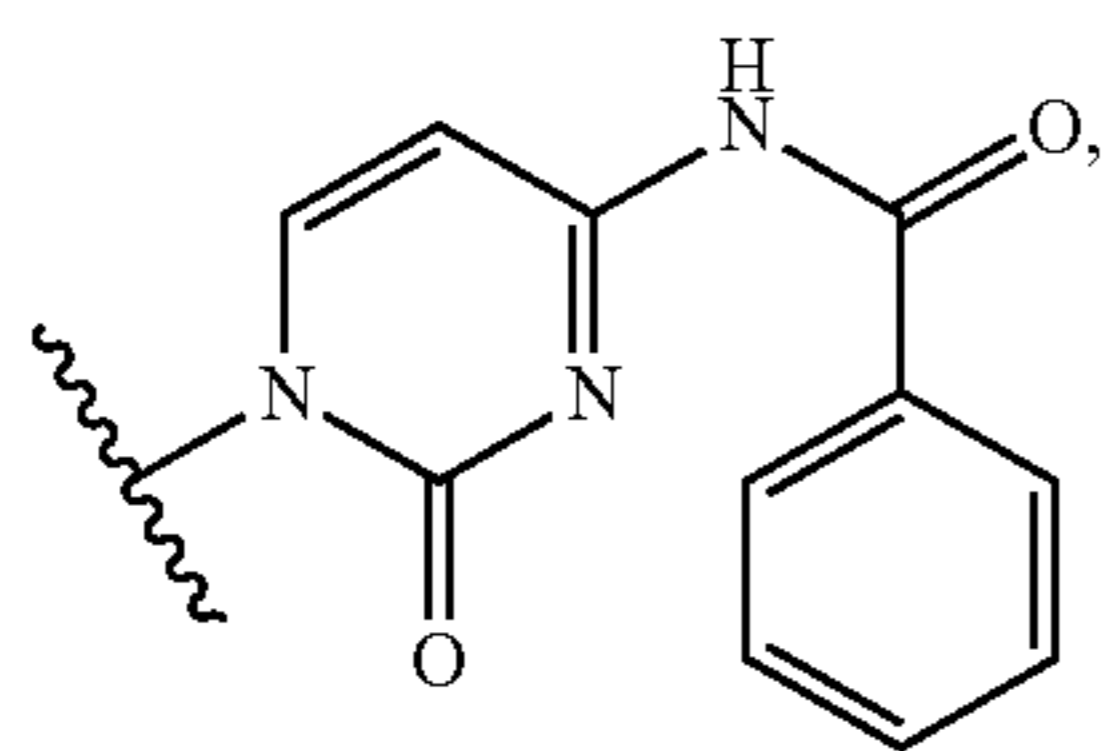
133

134

(i) contacting the compound of Formula (X) with a cleaving agent to form a compound of Formula (XI):



wherein R² is, independently for each occurrence, selected from the group consisting of:



and

wherein R² is at each position from 1 to 30 and 5' to 3':

Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC
8	PA
9	T
10	PC
11	PA

135

-continued

Position No. 5' to 3'	R ²
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG
18	PA
19	T
20	DPG
21	DPG
22	PC
23	PA
24	T
25	T
26	T
27	PC
28	T
29	PA
30	DPG

and

(j) contacting the compound of Formula (XI) with a deprotecting agent to form the oligomeric compound of Formula (E).

In an embodiment, step (d), step (f), step g(2), or combinations thereof further comprises contacting the compound of Formula (IV), Formula (VI), or the compound formed by the immediately prior step, respectively, with a capping agent.

In certain embodiments, each of step (d), step (f) and step g(2) further comprises contacting the compound of Formula (IV), Formula (VI), or the compound formed by the immediately prior step, respectively, with a capping agent.

In another embodiment, each step is performed in the presence of at least one solvent.

In yet another embodiment, the deblocking agent used in each step is a solution comprising a halogenated acid.

In still another embodiment, the deblocking agent used in each step is cyanoacetic acid.

In another embodiment, the halogenated acid is selected from the group consisting of chloroacetic acid, dichloroacetic acid, trichloroacetic acid, fluoroacetic acid, difluoroacetic acid, and trifluoroacetic acid.

In yet another embodiment, the halogenated acid is trifluoroacetic acid.

In still another embodiment, at least one of steps (c), (e1), and (f) further comprise the step of contacting the deblocked compound of each step with a neutralization agent.

In another embodiment, each of steps (c), (e1), and (f) further comprise the step of contacting the deblocked compound of each step with a neutralization agent.

In yet another embodiment, the neutralization agent is in a solution comprising dichloromethane and isopropyl alcohol.

136

In still another embodiment, the neutralization agent is a monoalkyl, dialkyl, or trialkyl amine.

In another embodiment, the neutralization agent is N,N-diisopropylethylamine.

In yet another embodiment, the deblocking agent used in each step is a solution comprising 4-cyanopyridine, dichloromethane, trifluoroacetic acid, trifluoroethanol, and water.

In still another embodiment, the capping agent is in a solution comprising ethylmorpholine and methylpyrrolidinone.

In another embodiment, the capping agent is an acid anhydride.

In yet another embodiment, the acid anhydride is benzoic anhydride.

In still another embodiment, the compound of Formula (VIII), compound (C), and compound (F) are each, independently, in a solution comprising ethylmorpholine and dimethylimidazolidinone.

In another embodiment, the cleavage agent comprises dithiothreitol and 1,8-diazabicyclo[5.4.0]undec-7-ene.

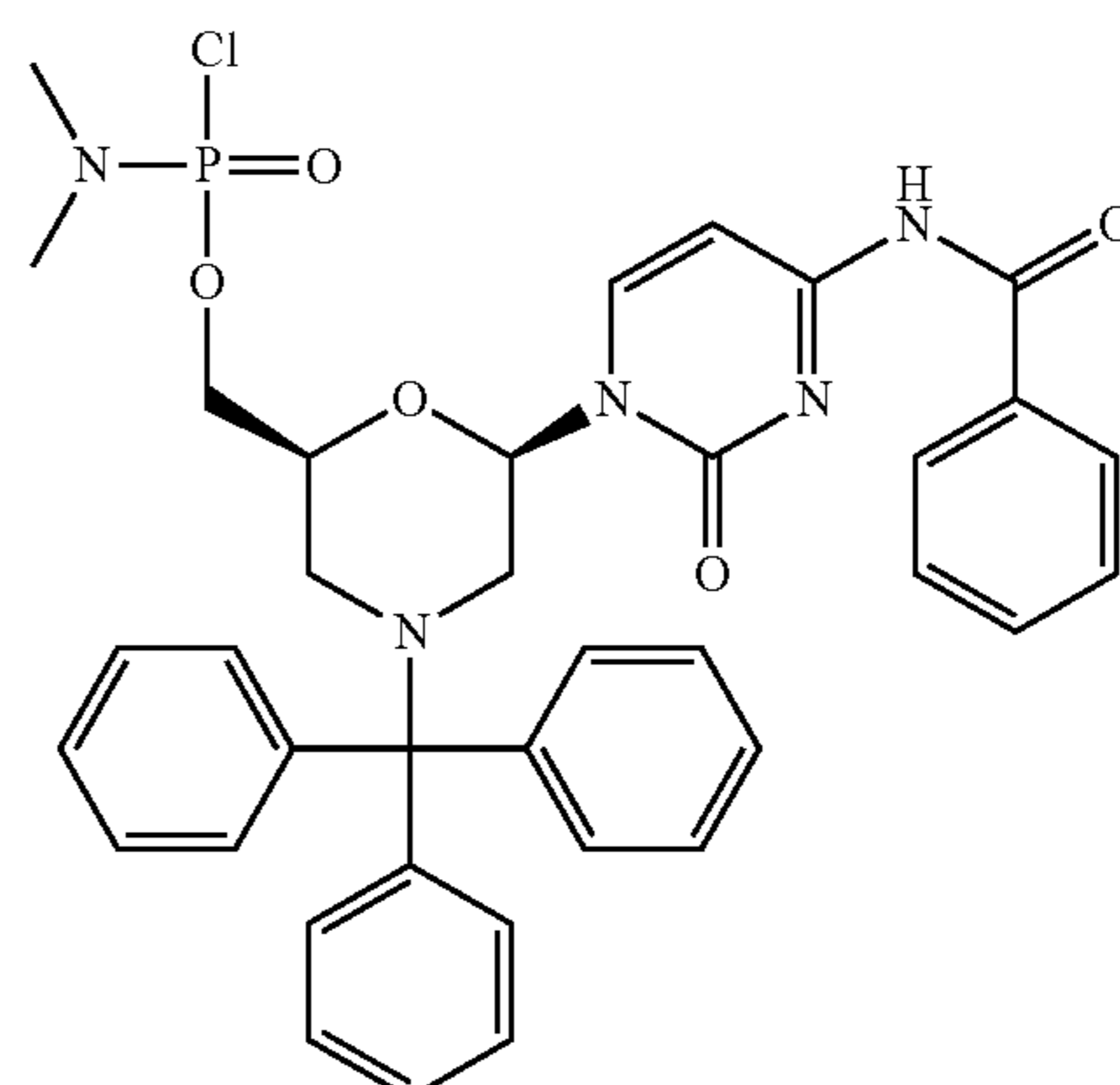
In yet another embodiment, the cleavage agent is in a solution comprising N-methyl-2-pyrrolidone.

In still another embodiment, the deprotecting agent comprises NH₃.

In another embodiment, the deprotecting agent is in an aqueous solution.

In yet another embodiment, the support-medium comprises polystyrene with 1% crosslinked divinylbenzene.

In another embodiment, the compound of Formula (C) is of Formula (C1):



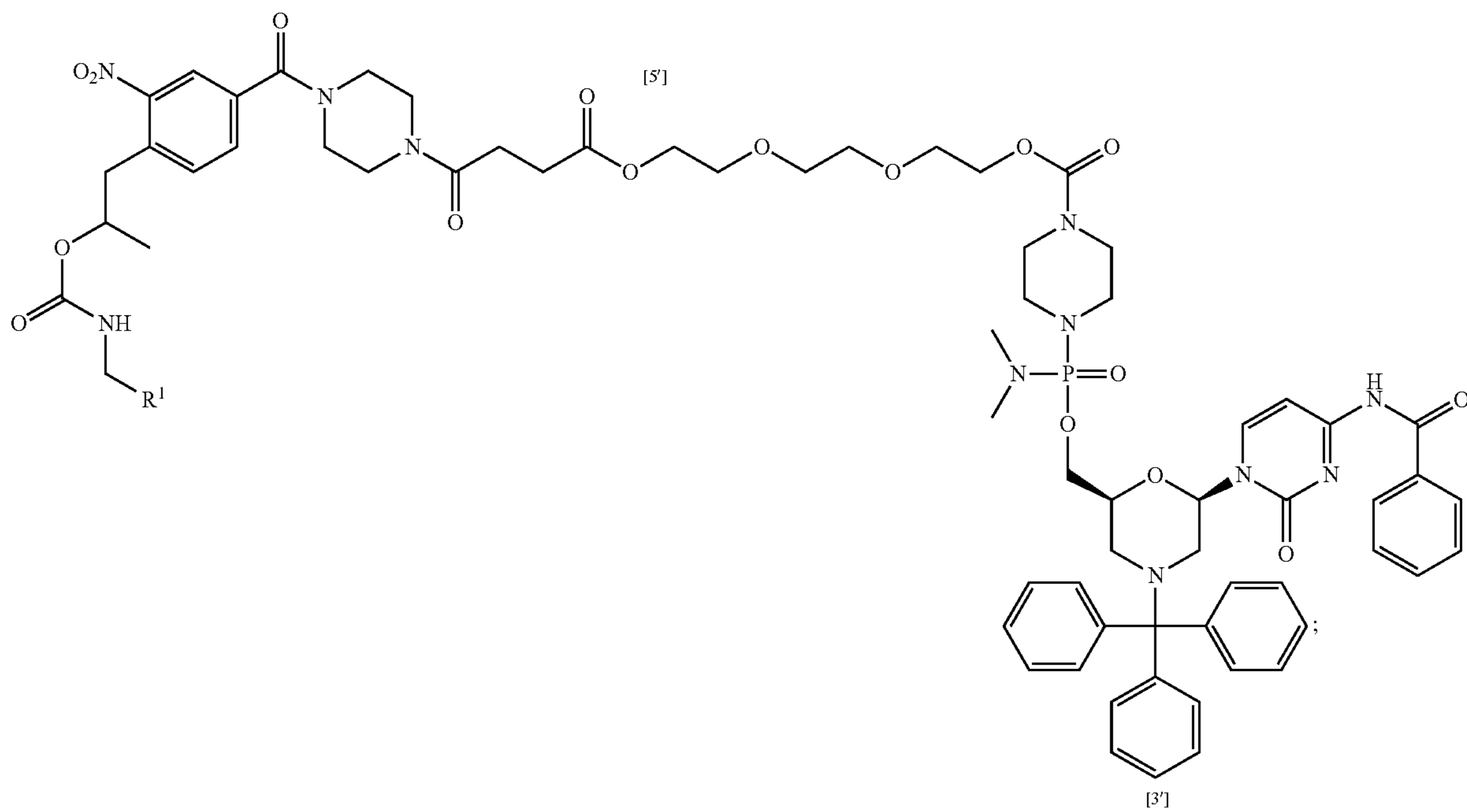
(C1)

137

In another embodiment, the compound of Formula (V) is of Formula (Va):

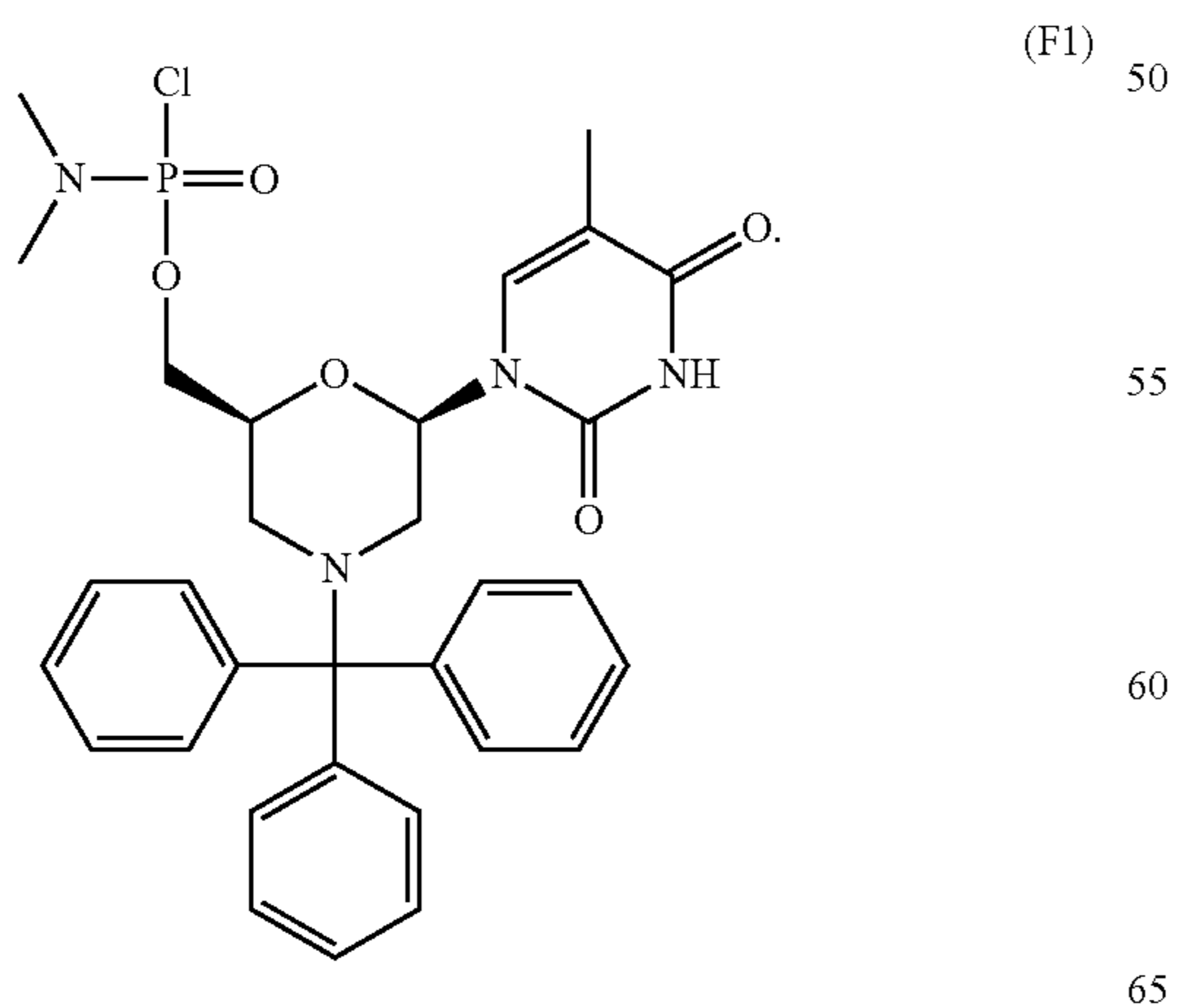
138

(Va)



wherein R^1 is a support-medium.

In another embodiment, the compound of Formula (F) is of Formula (F1):



(F1) 50

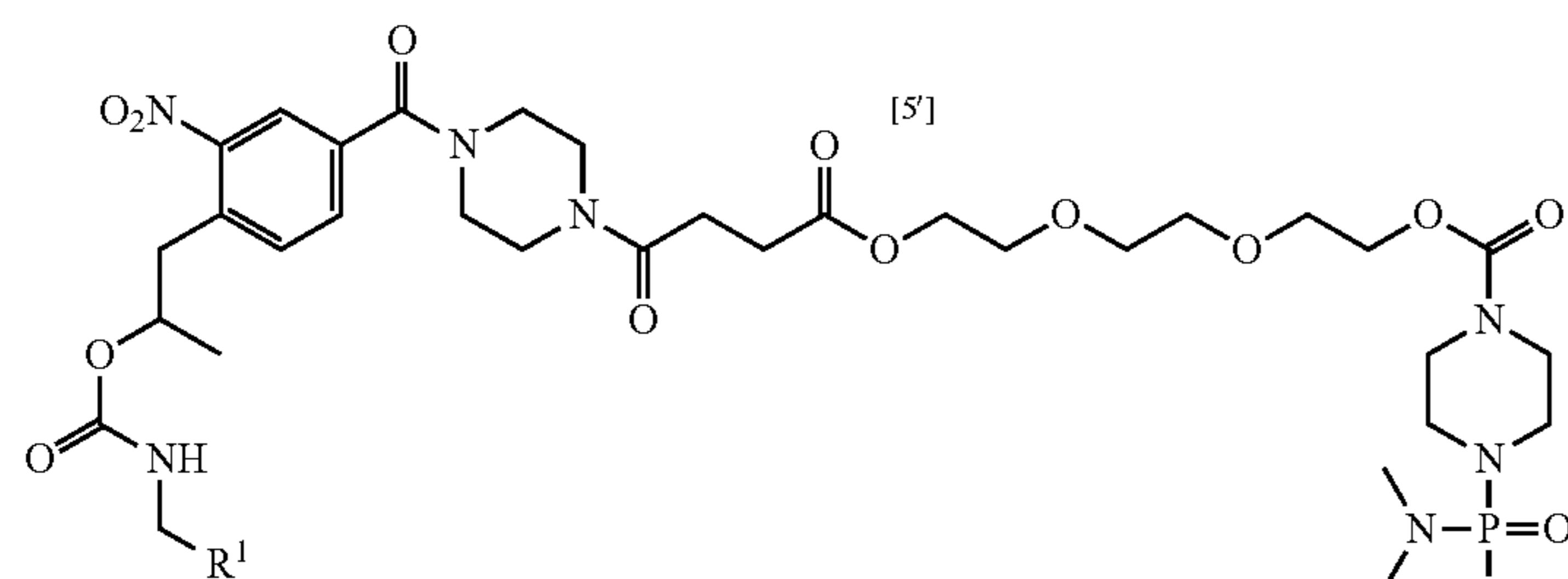
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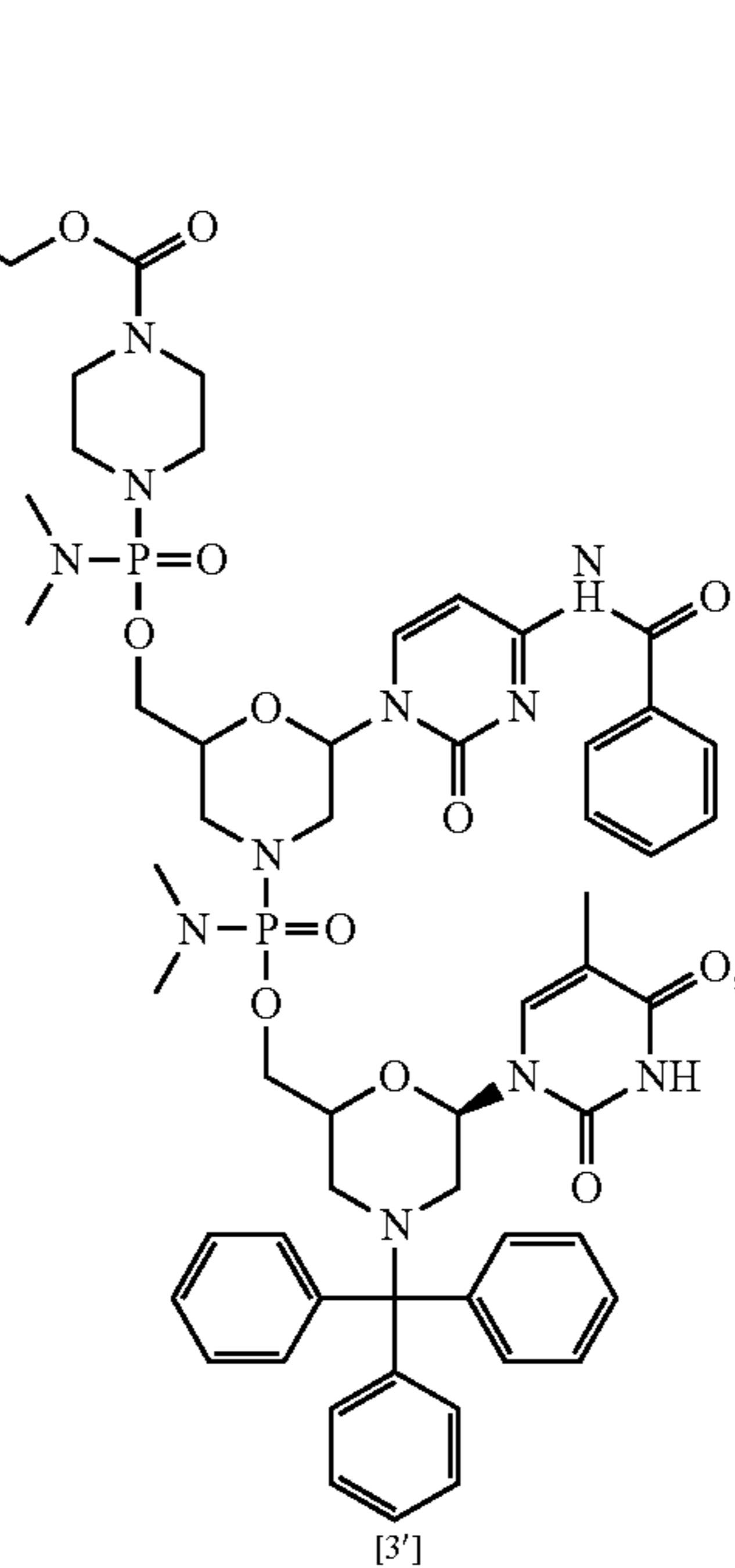
139

In another embodiment, the compound of Formula (VII) is of Formula (VIIa):



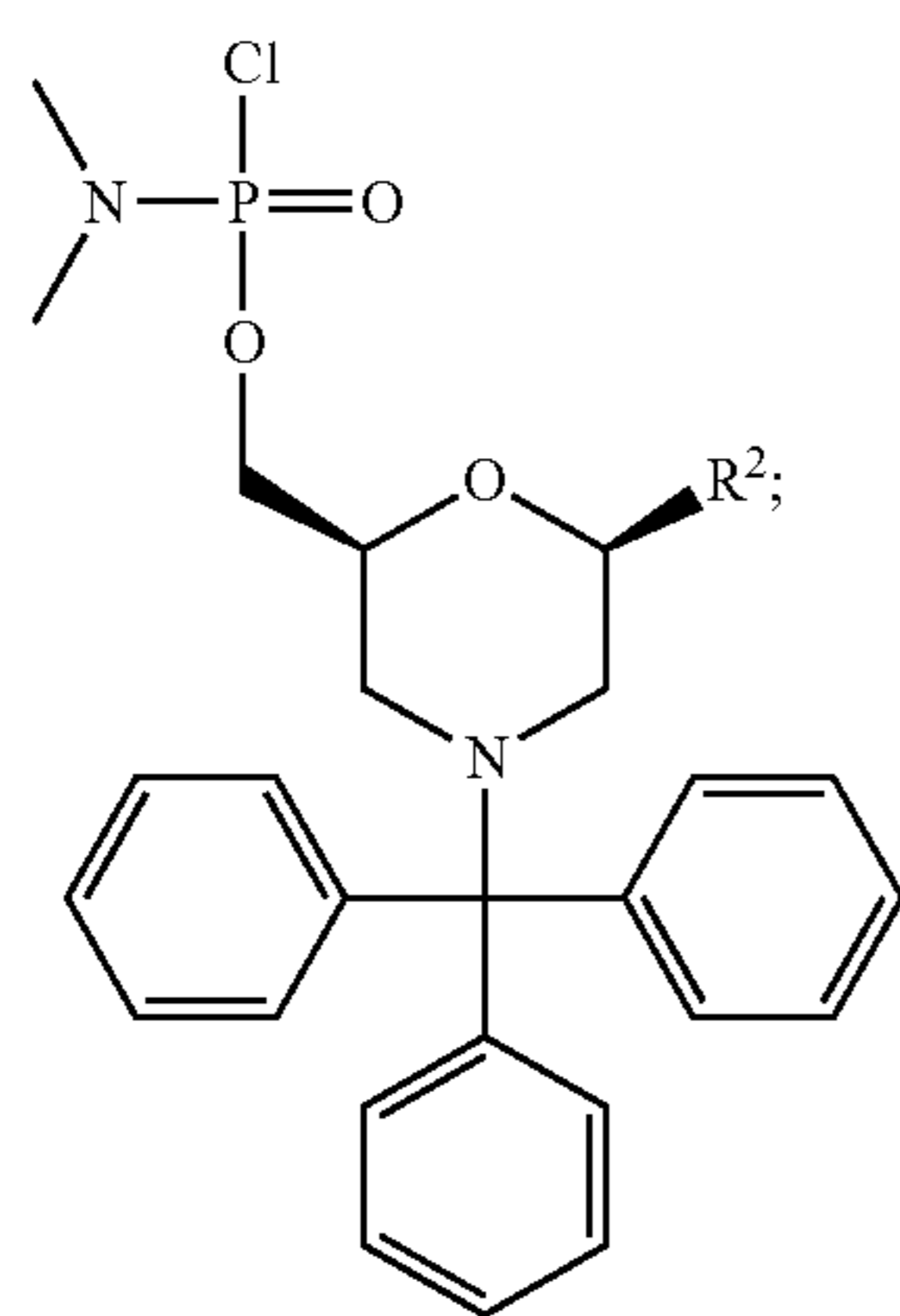
140

(VIIa)

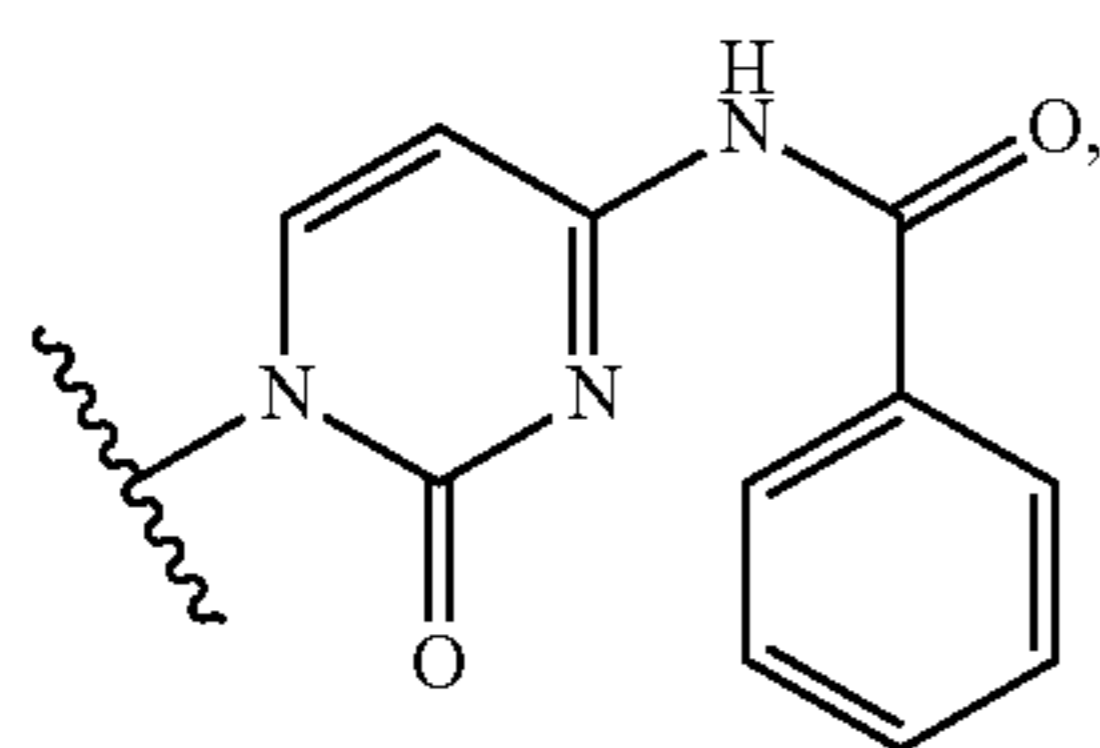


wherein R¹ is a support-medium.

In another embodiment, the compound of Formula (VIII) is of Formula (VIIIa):



wherein R² is, independently for each compound of Formula (VIIIa), selected from the group consisting of:



(PC)

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-continued

(VIIIa)

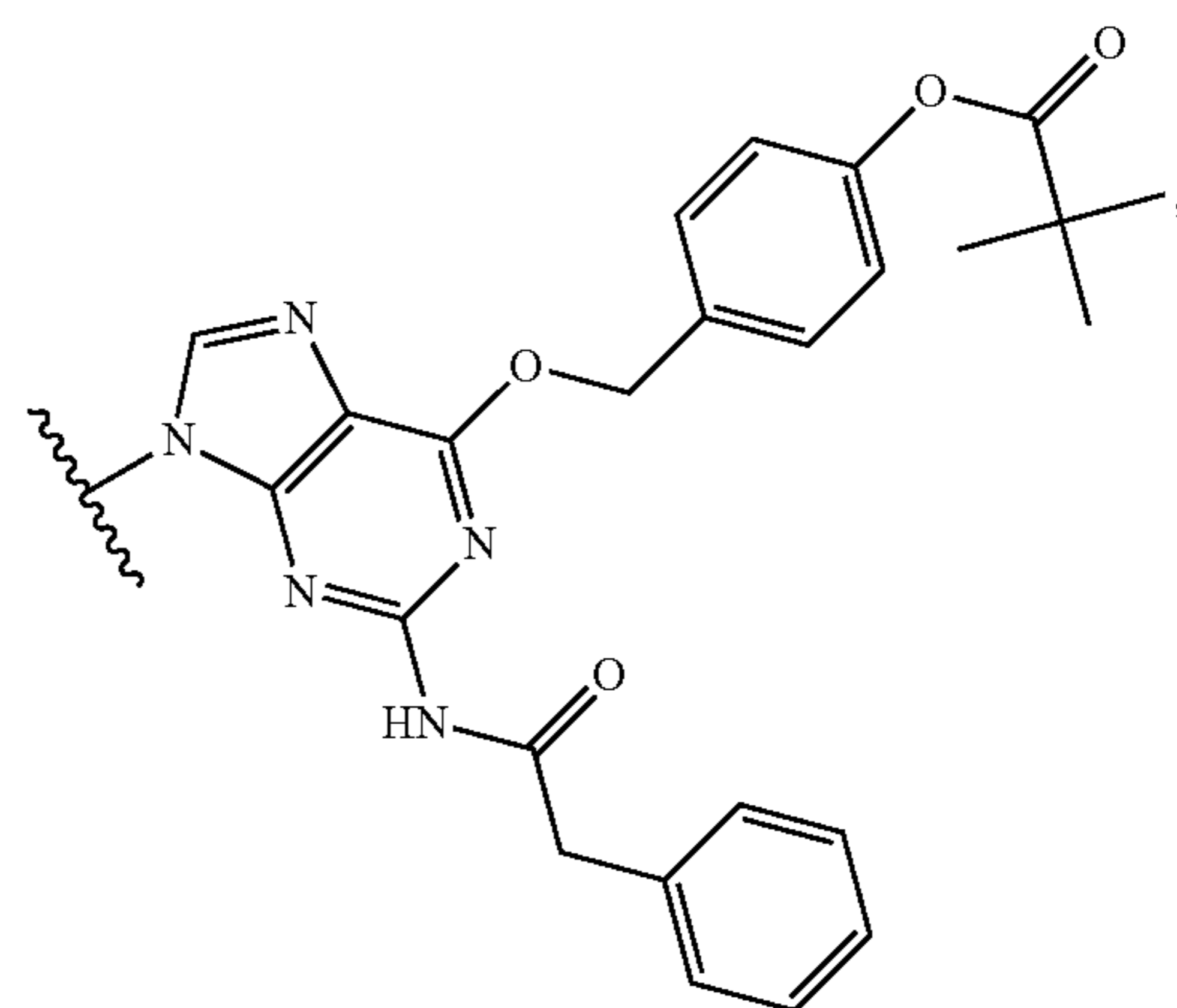
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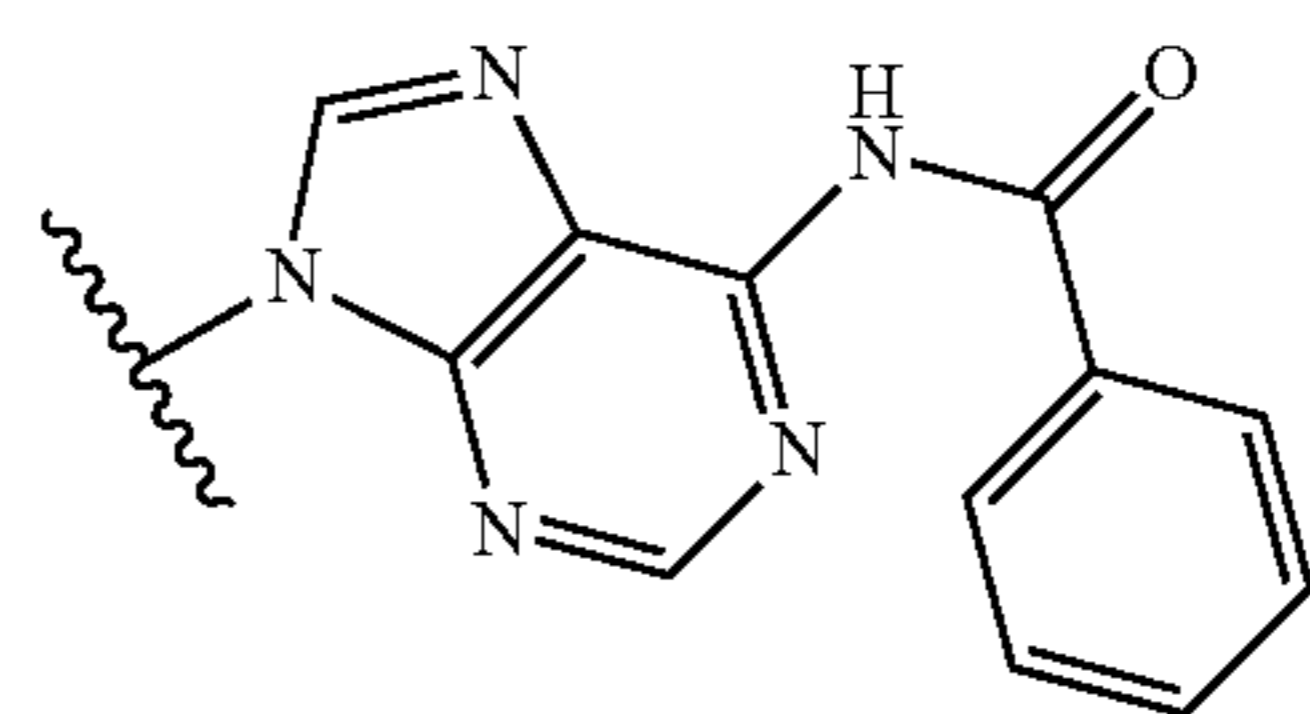
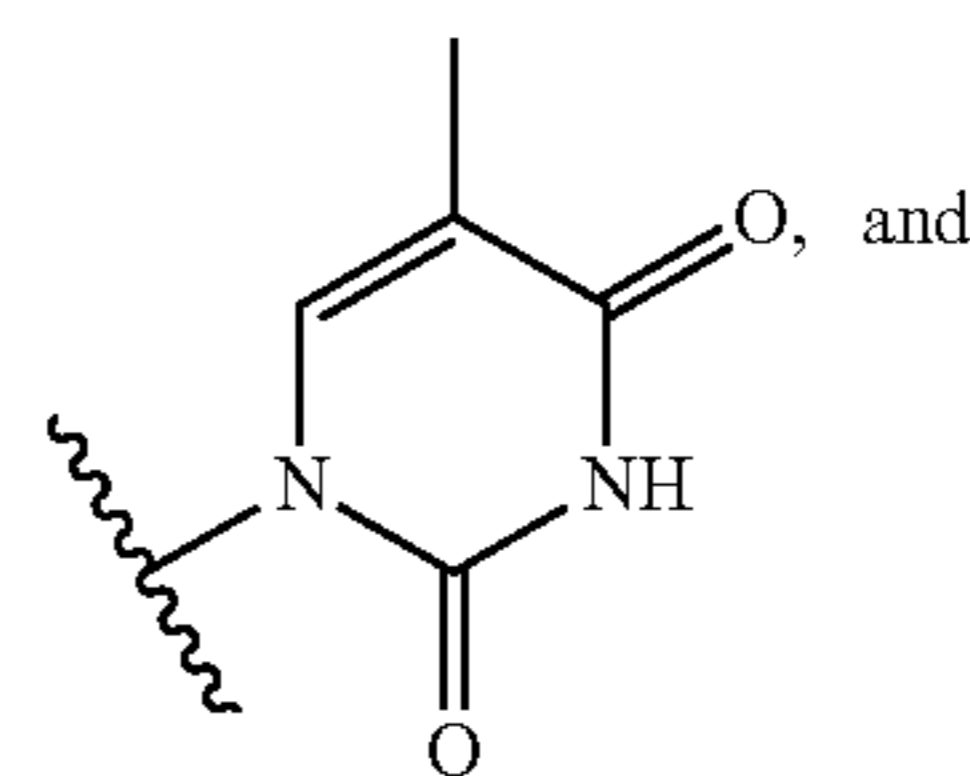
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(DPG)

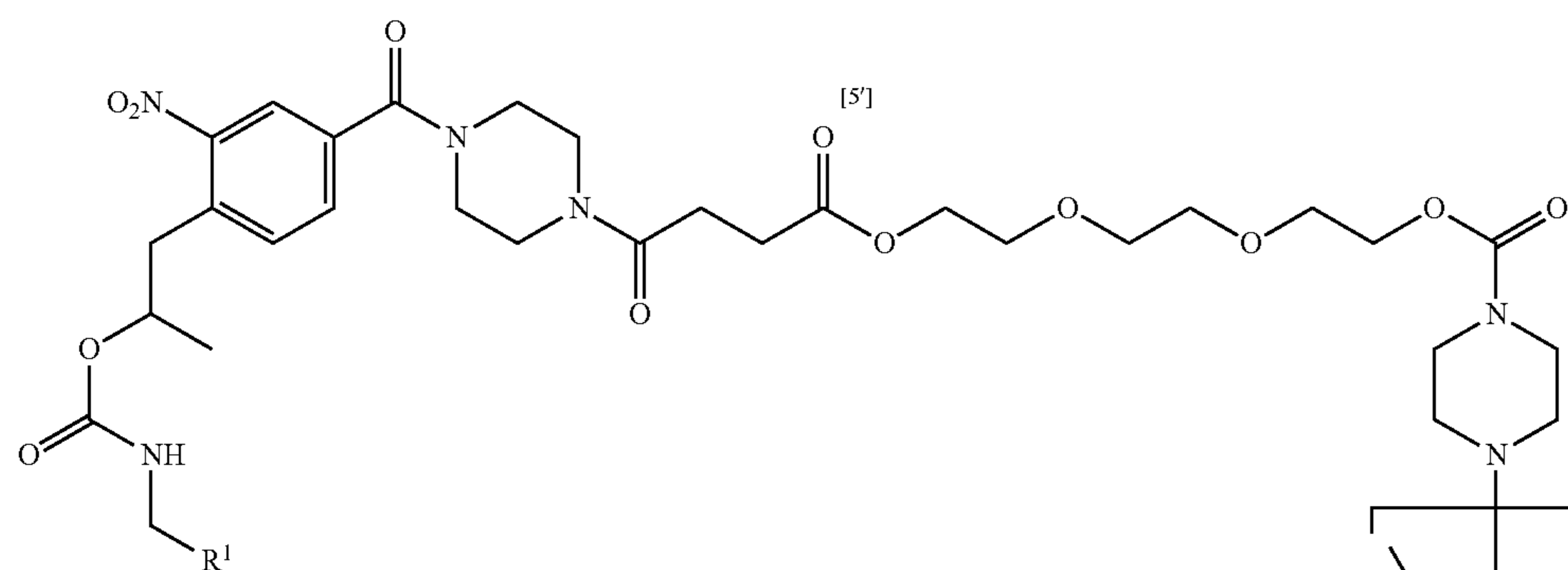
(T)



(PA)

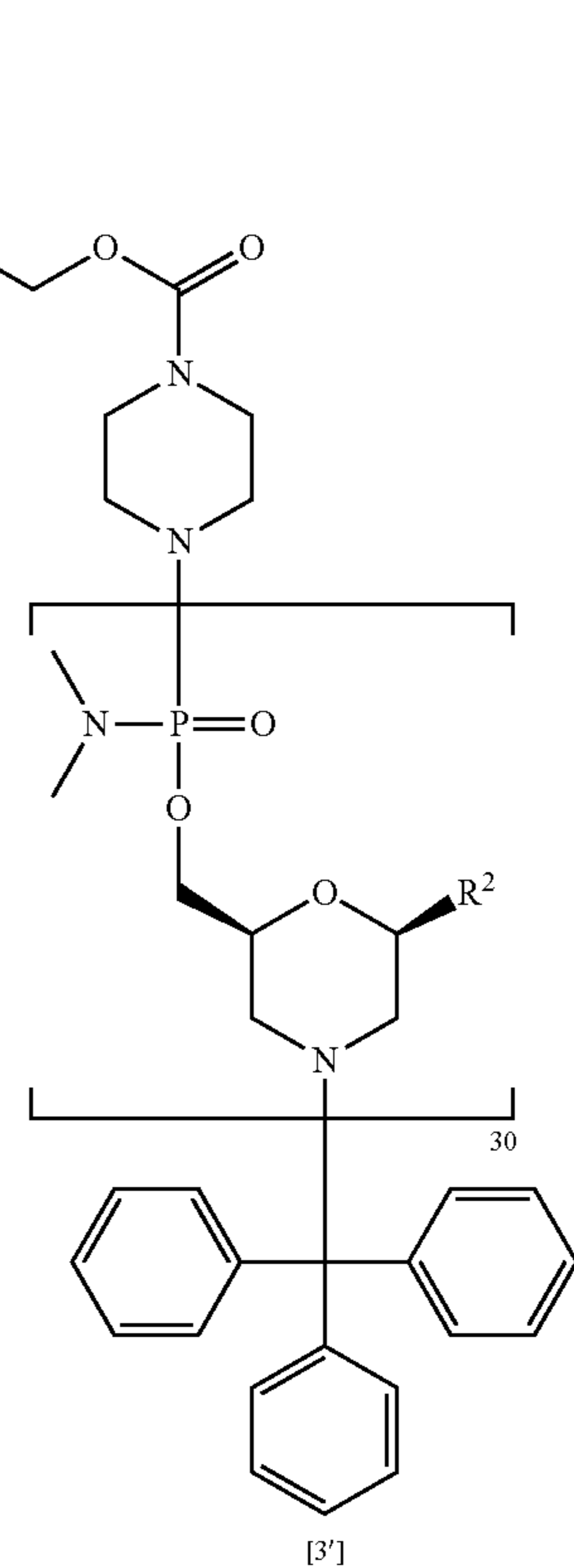
141

In another embodiment, the compound of Formula (IX) is of Formula (IXa):

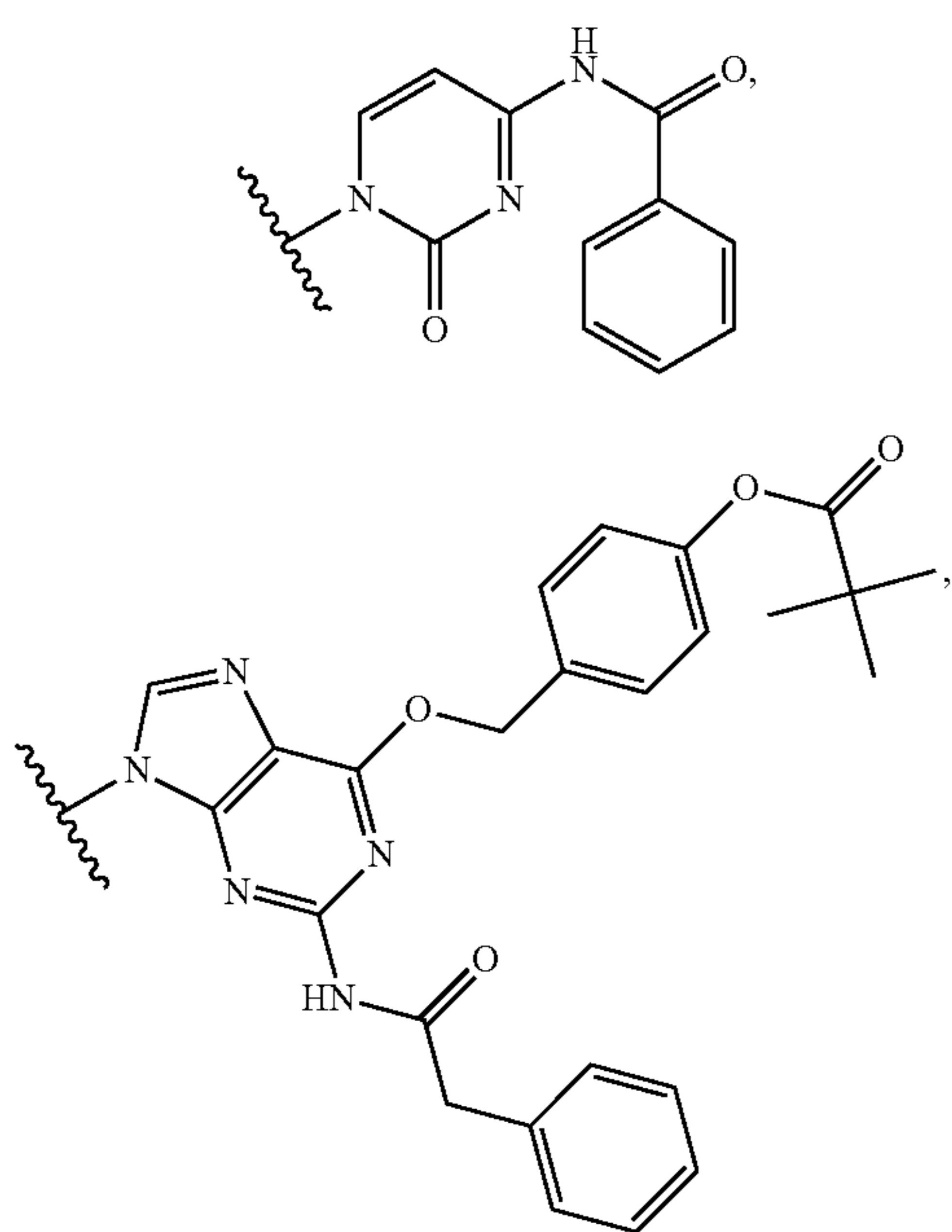


142

(IXa)

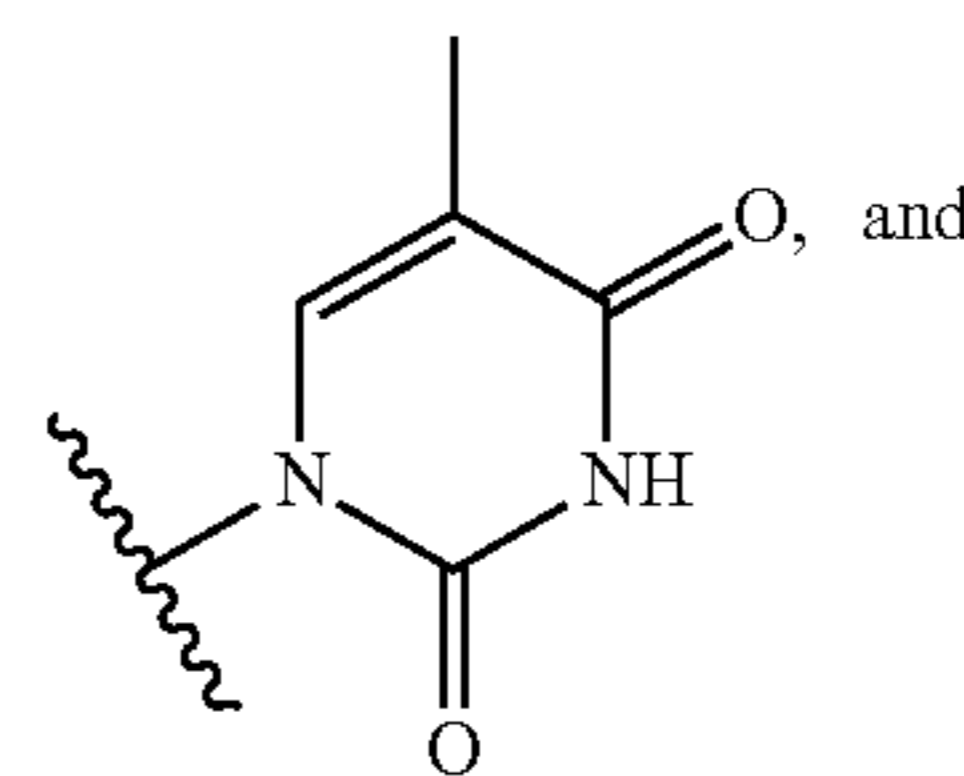


or a pharmaceutically acceptable salt thereof, wherein R¹ is a support-medium, and R² is, independently at each occurrence, selected from the group consisting of:

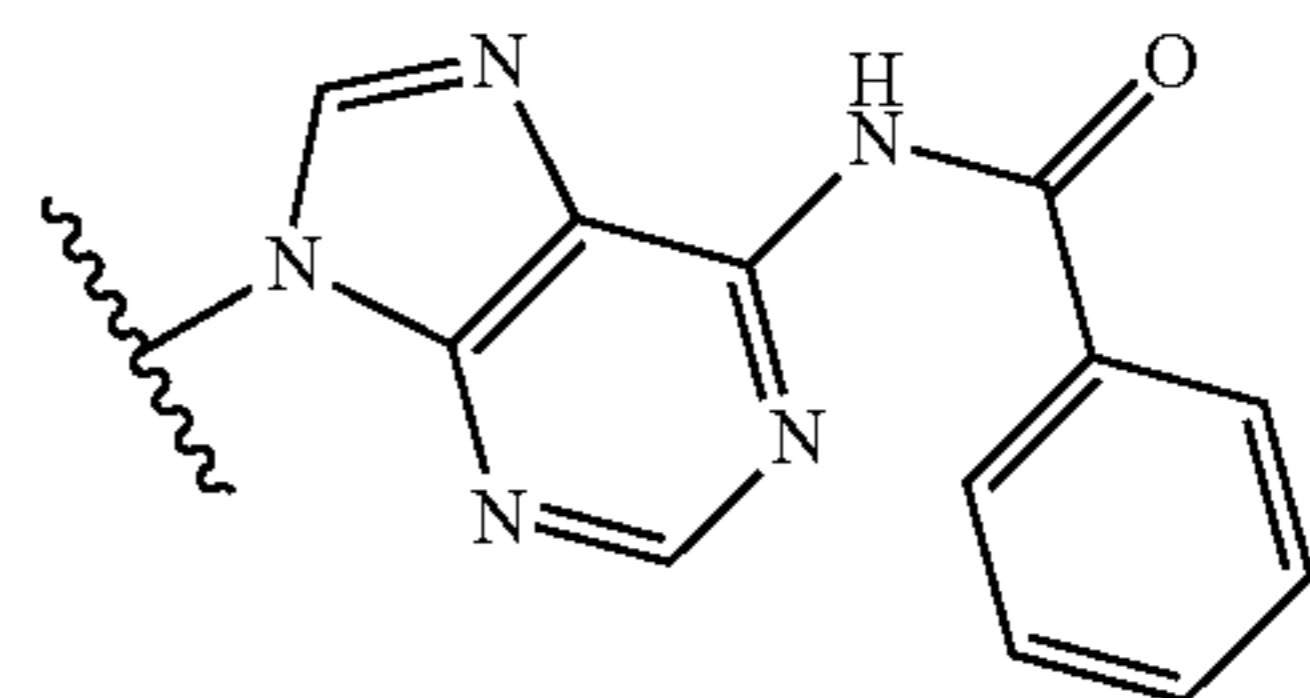


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(T)



(PA)



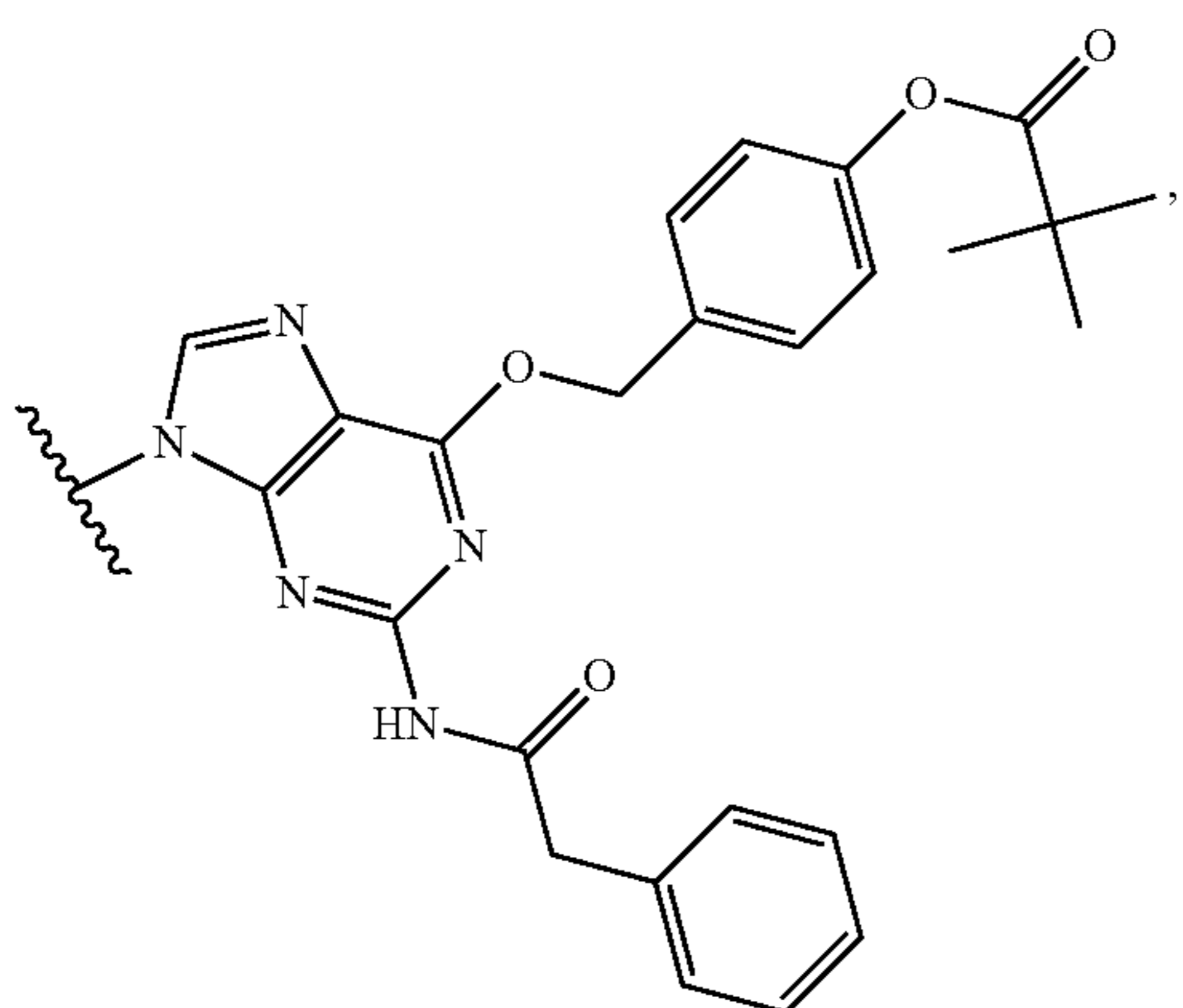
and

wherein R² is at each position from 1 to 30 and 5' to 3':

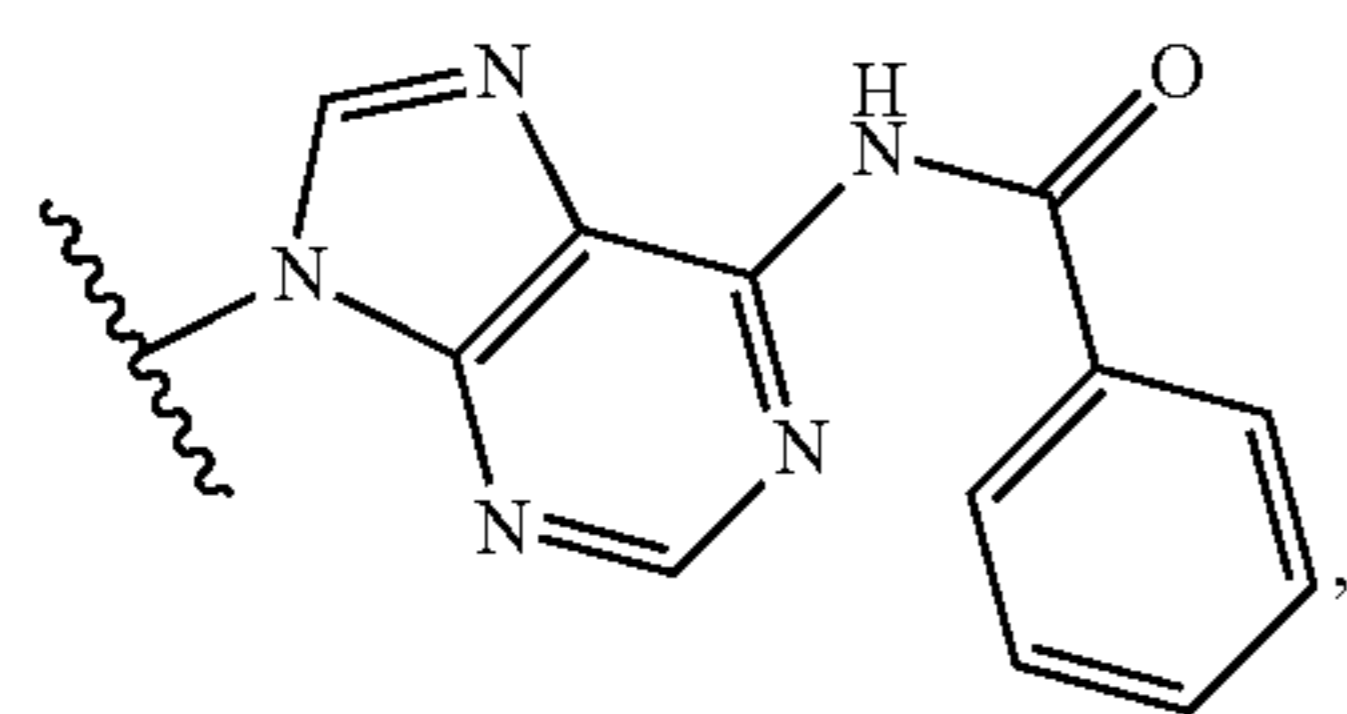
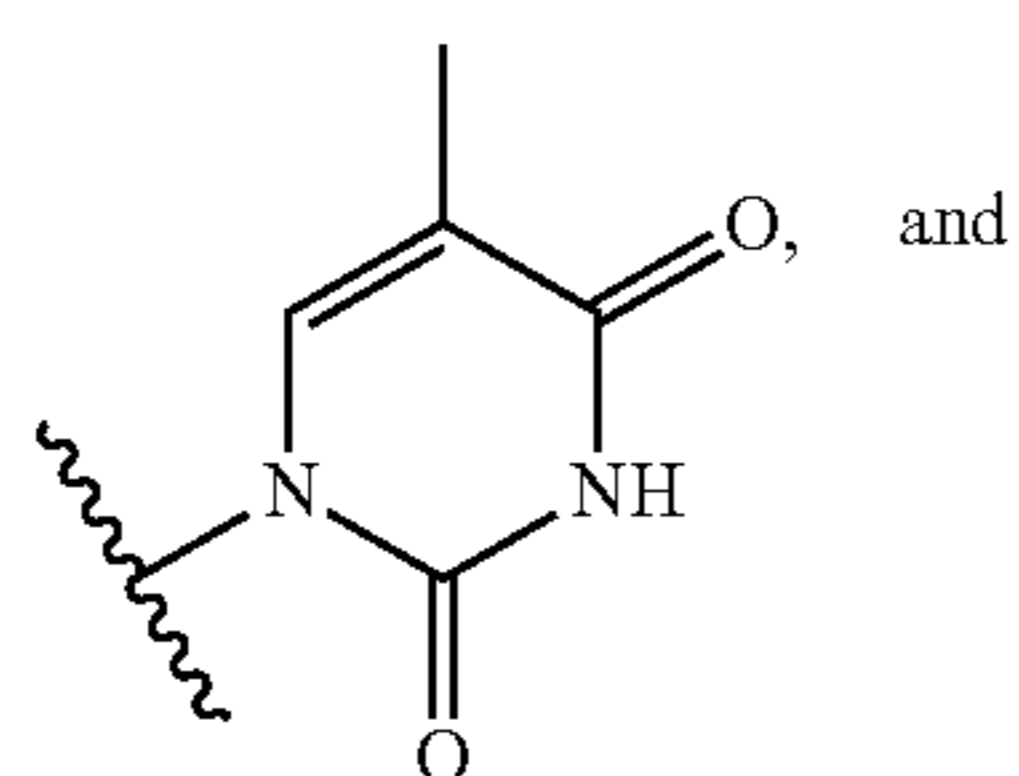
Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC
8	PA
9	T
10	PC
11	PA

145

-continued



(DPG)



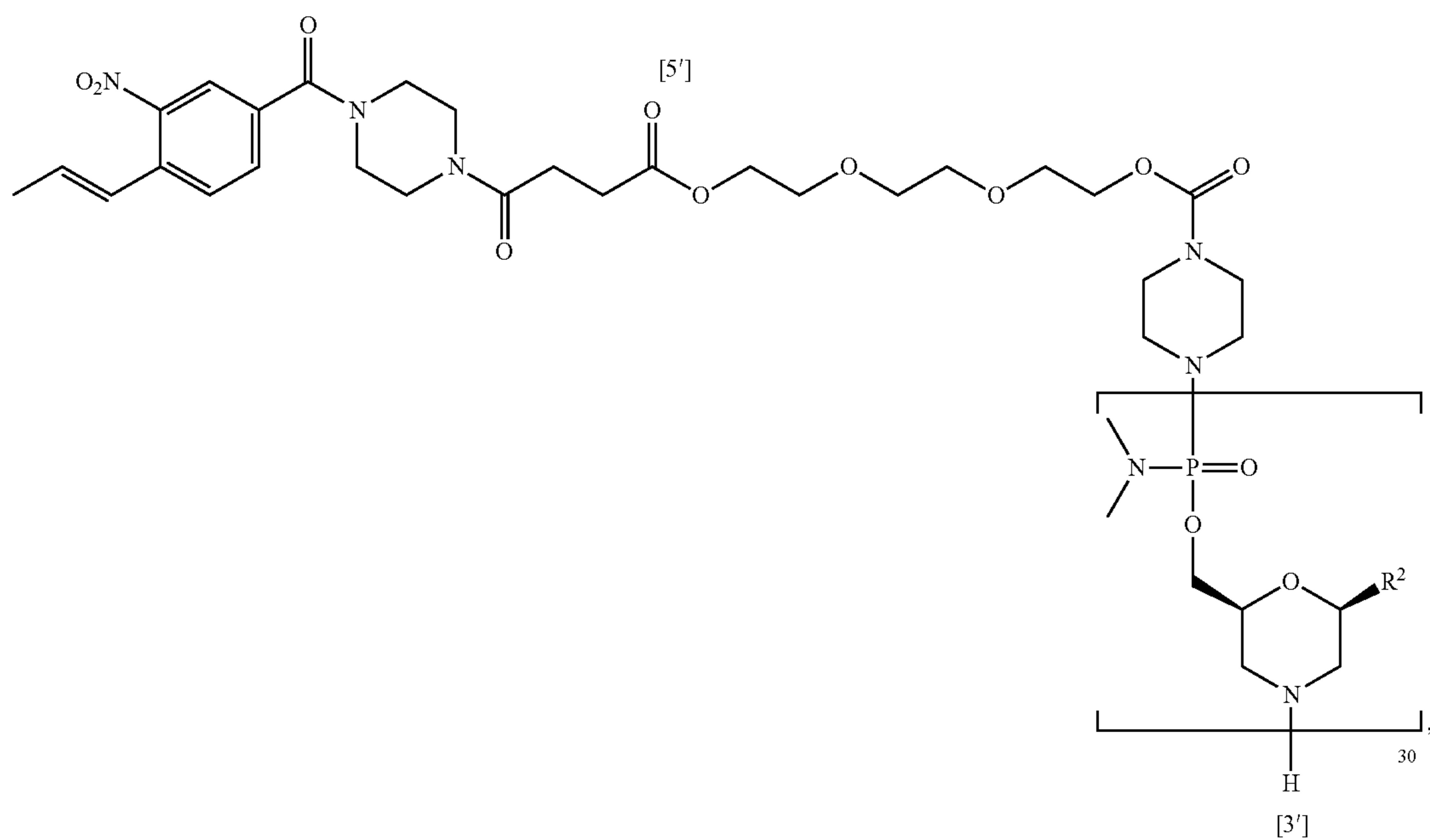
146

and

wherein R² is at each position from 1 to 30 and 5' to 3':

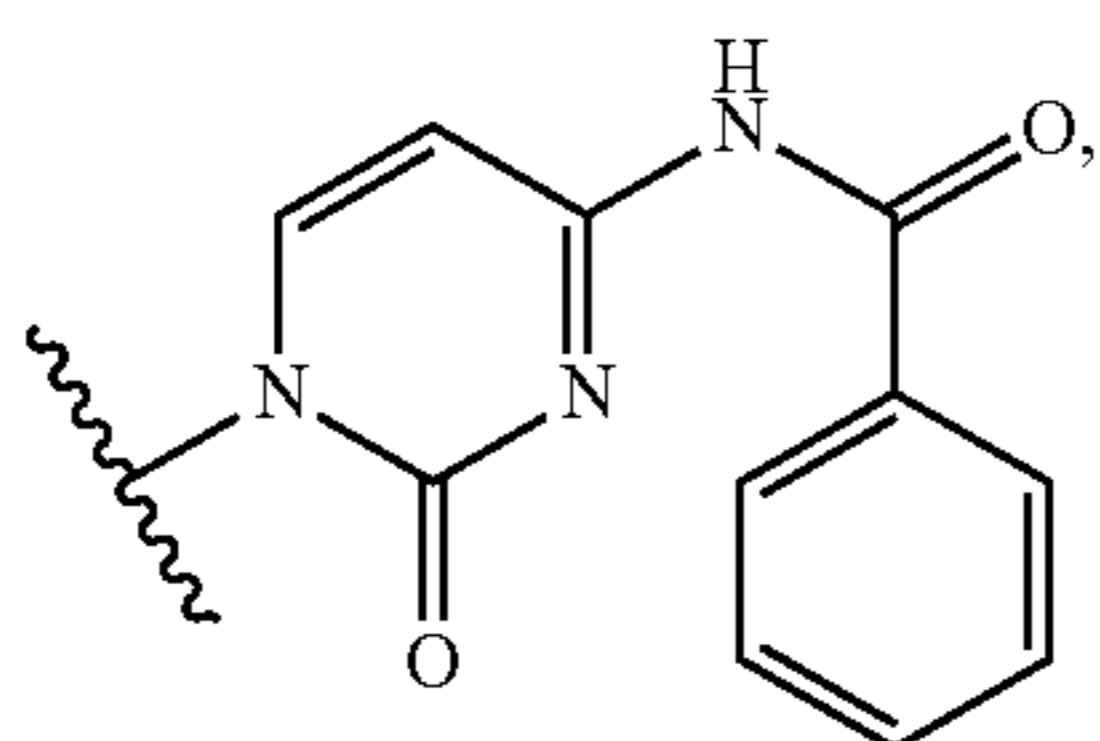
	Position No. 5' to 3'	R ²
5	1	PC
	2	T
	3	PC
10	4	PC
	5	PA
	6	PA
15	7	PC
	8	PA
	9	T
20	10	PC
	11	PA
	12	PA
25	13	DPG
	14	DPG
	15	PA
30	16	PA
	17	DPG
	18	PA
30	19	T
	20	DPG
	21	DPG
30	22	PC
	23	PA
	24	T
30	25	T
	26	T
	27	PC
30	28	T
	29	PA
	30	DPG

In another embodiment, the compound of Formula (XI) is of Formula (XIa):



147

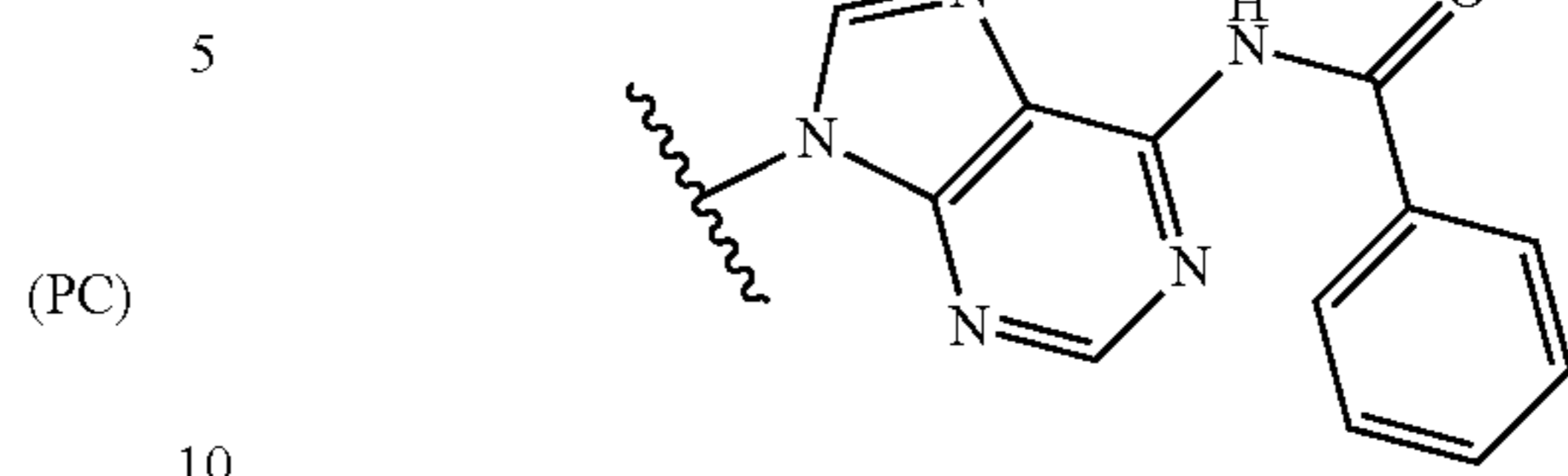
or a pharmaceutically acceptable salt thereof, wherein:
R² is, independently at each occurrence, selected from the group consisting of:



148

-continued

(PA)



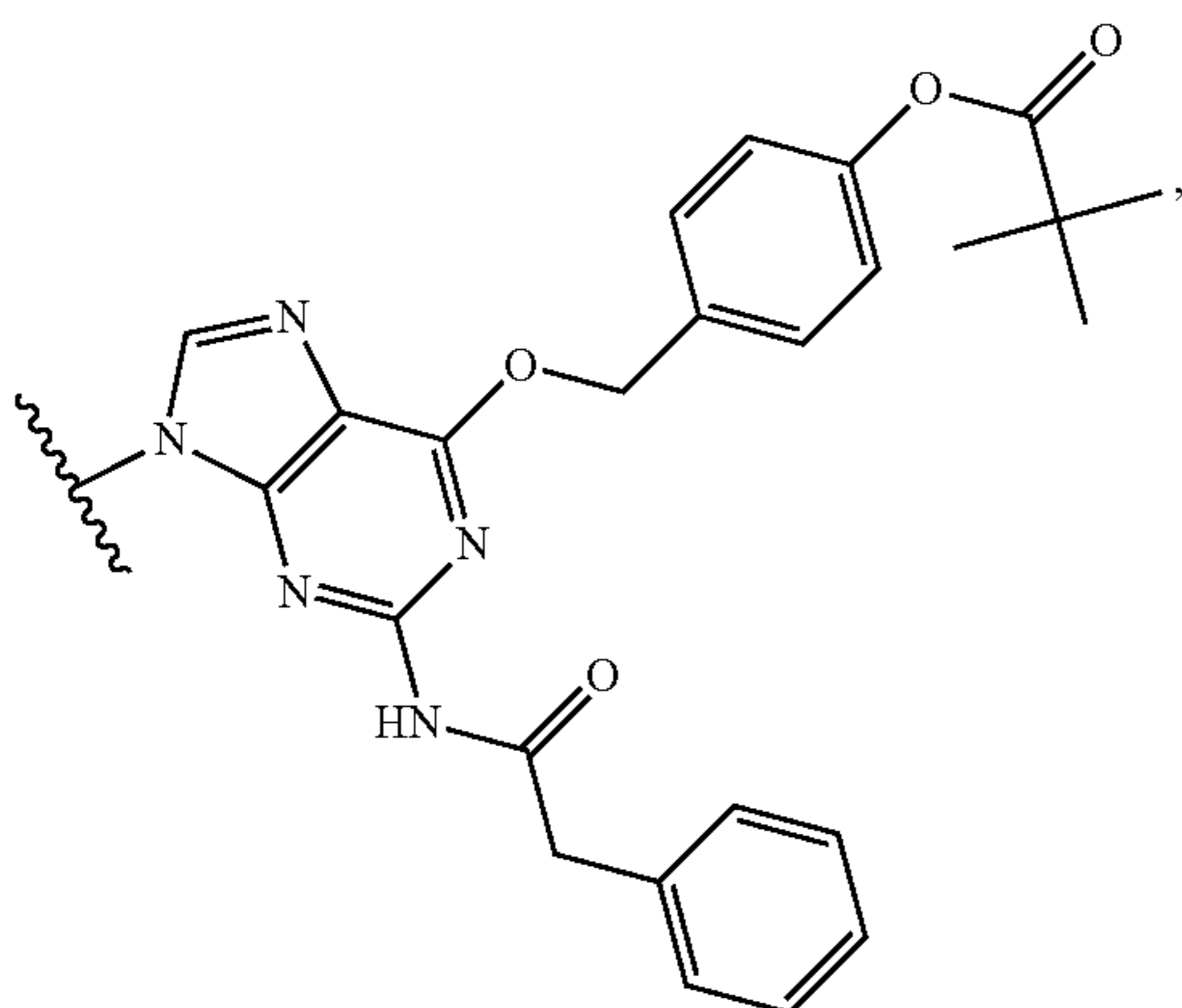
15 and

wherein R² is at each position from 1 to 30 and 5' to 3':

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(DPG)



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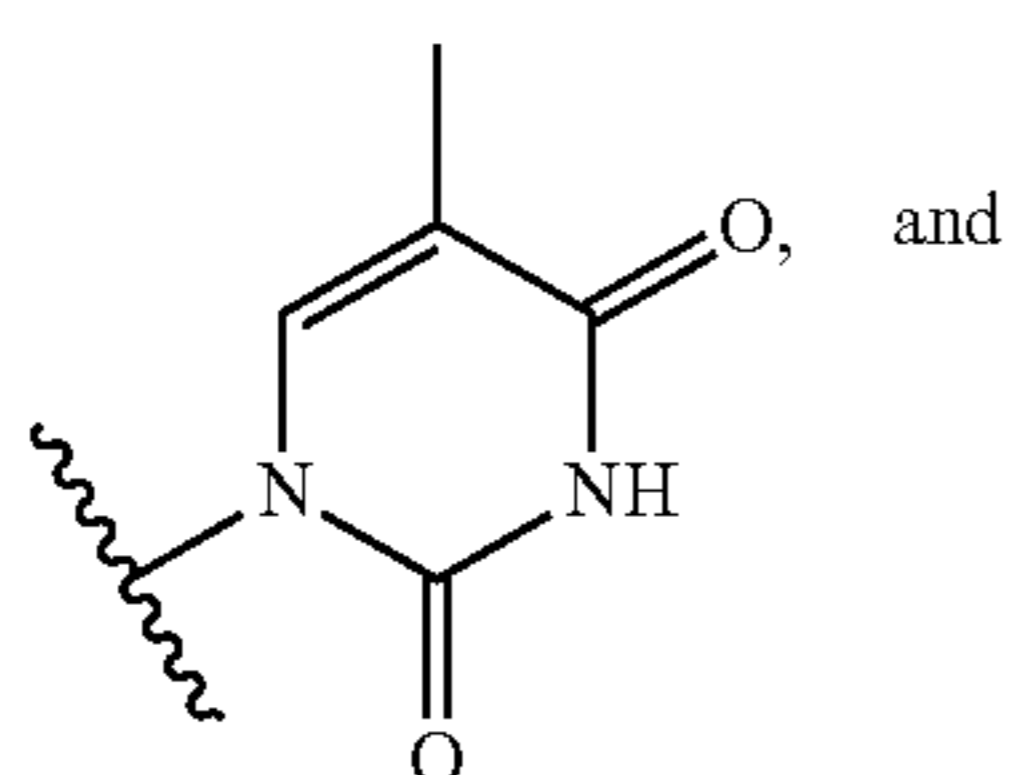
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(T)

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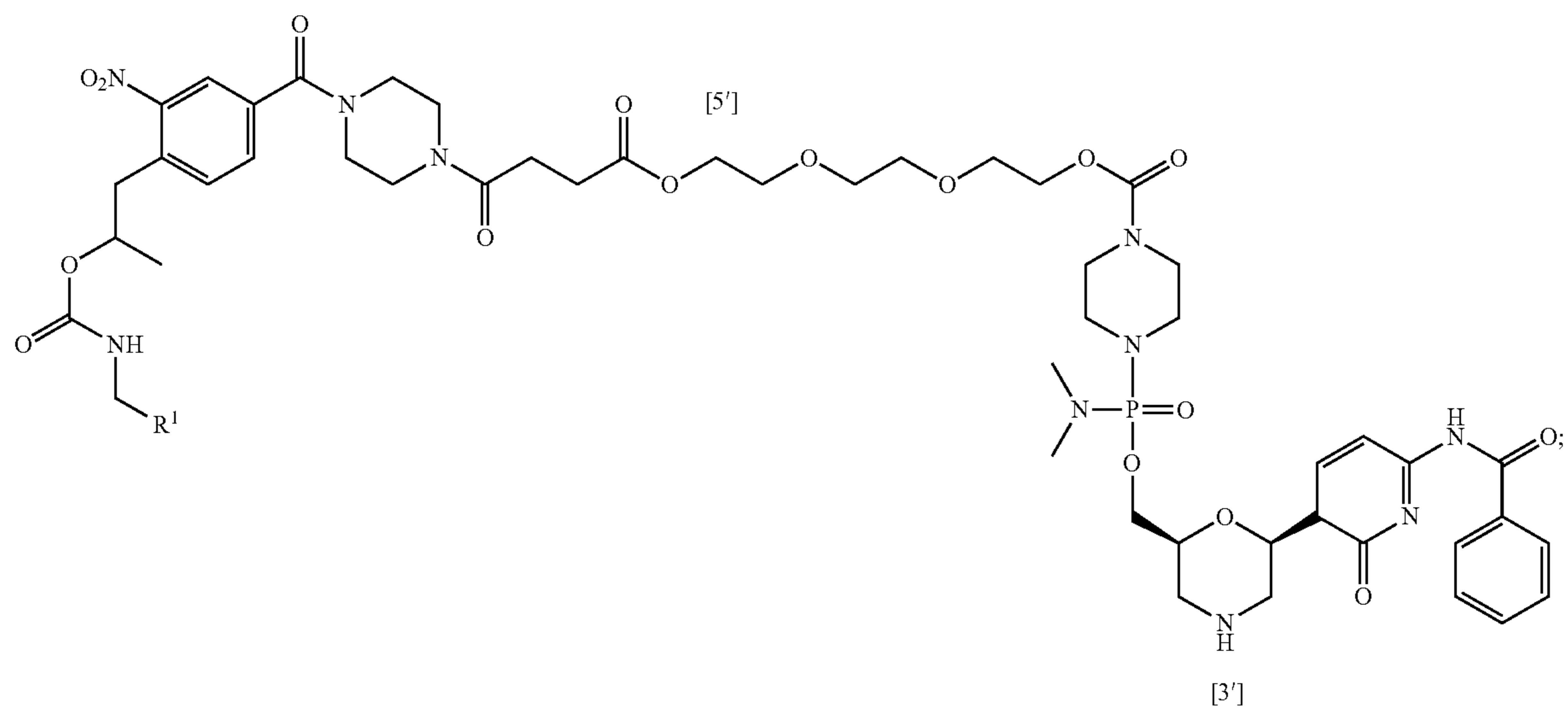
Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC
8	PA
9	T
10	PC
11	PA
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG
18	PA
19	T
20	DPG
21	DPG
22	PC
23	PA
24	T
25	T
26	T
27	PC
28	T
29	PA
30	DPG

149

In another embodiment, the compound of Formula (VI) is of Formula (VIa):

150

(VIa)



wherein R¹ is a support-medium.

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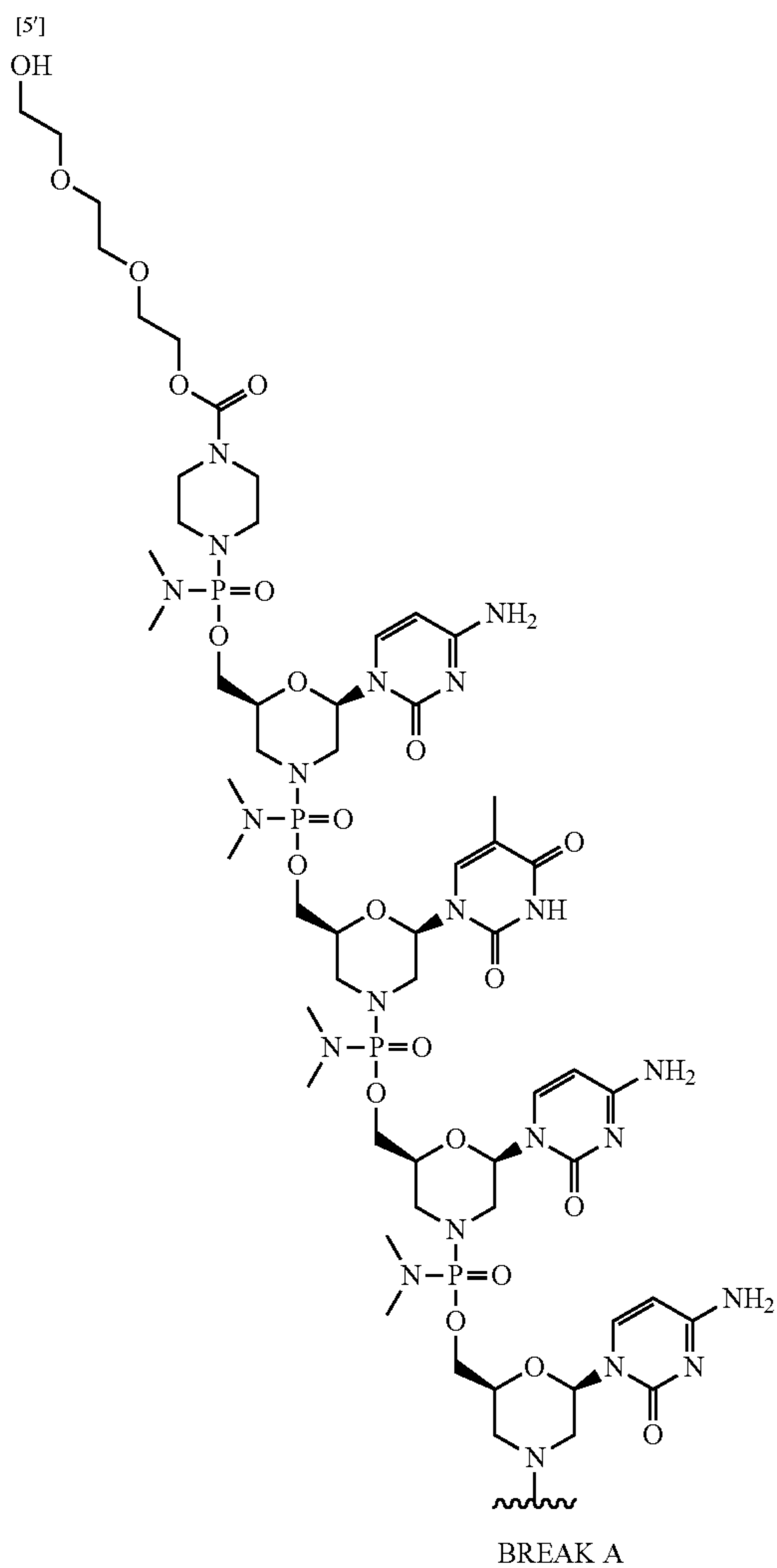
55

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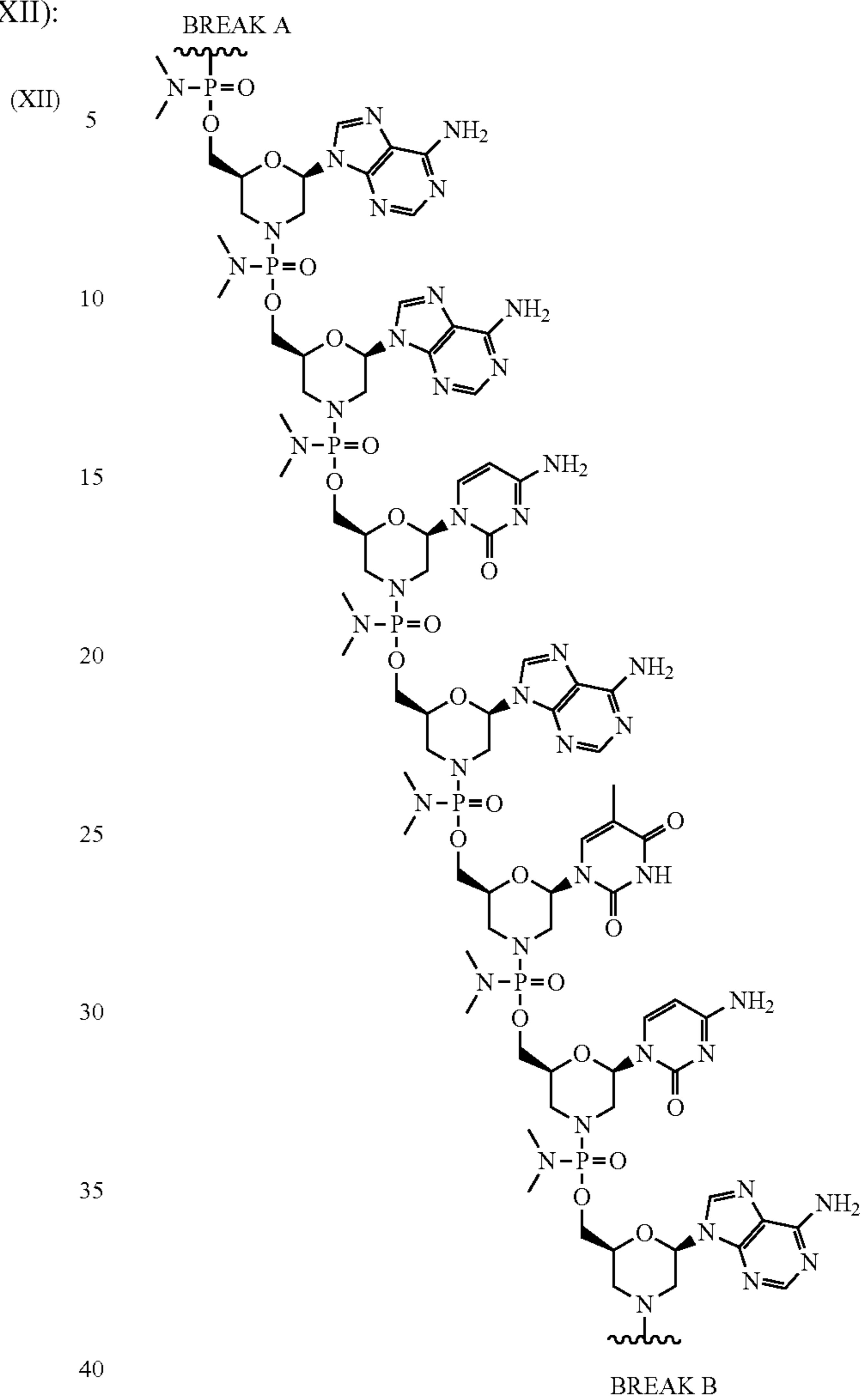
151

In still another embodiment, the oligomeric compound of Formula (E) is an oligomeric compound of Formula (XII):



152

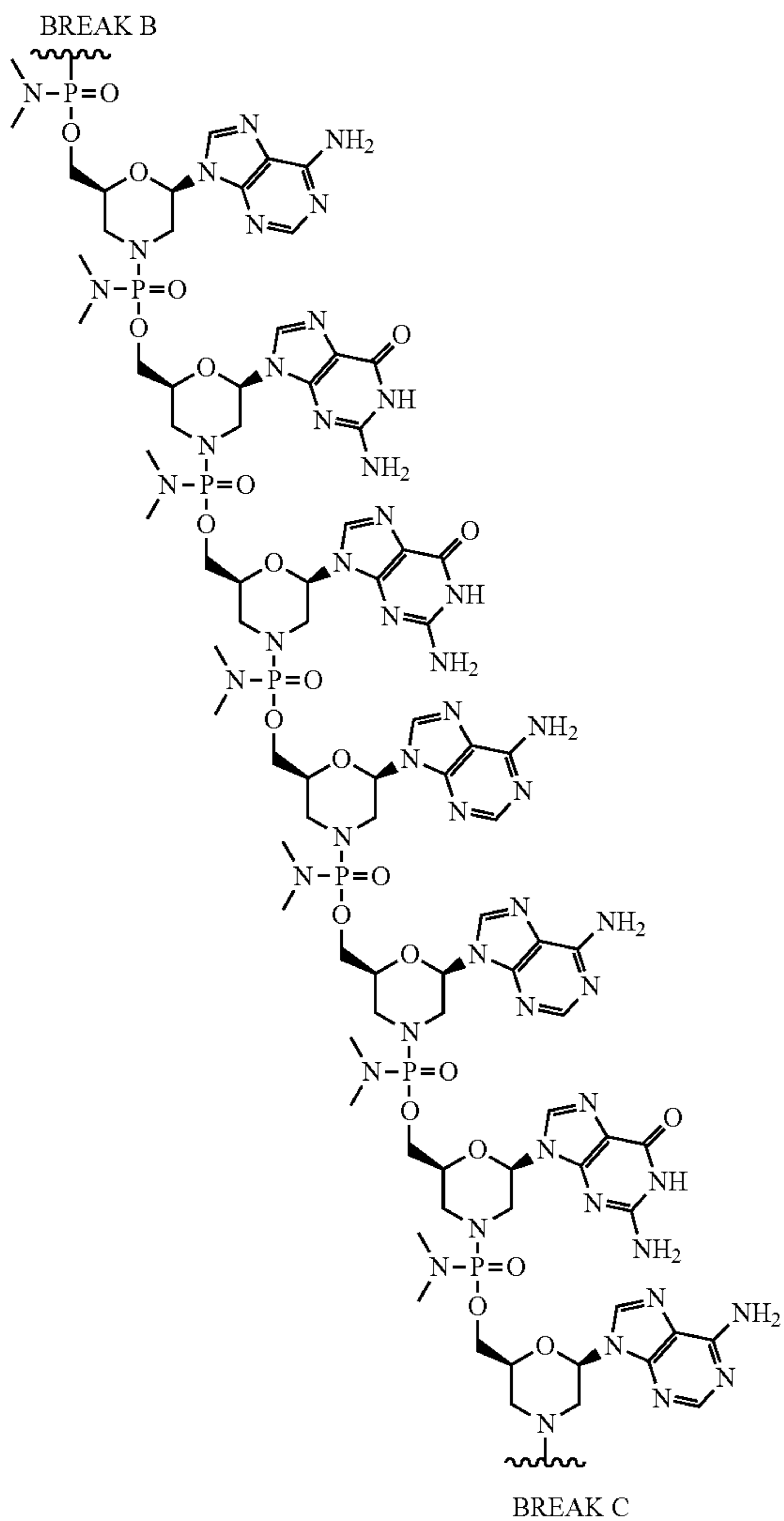
-continued



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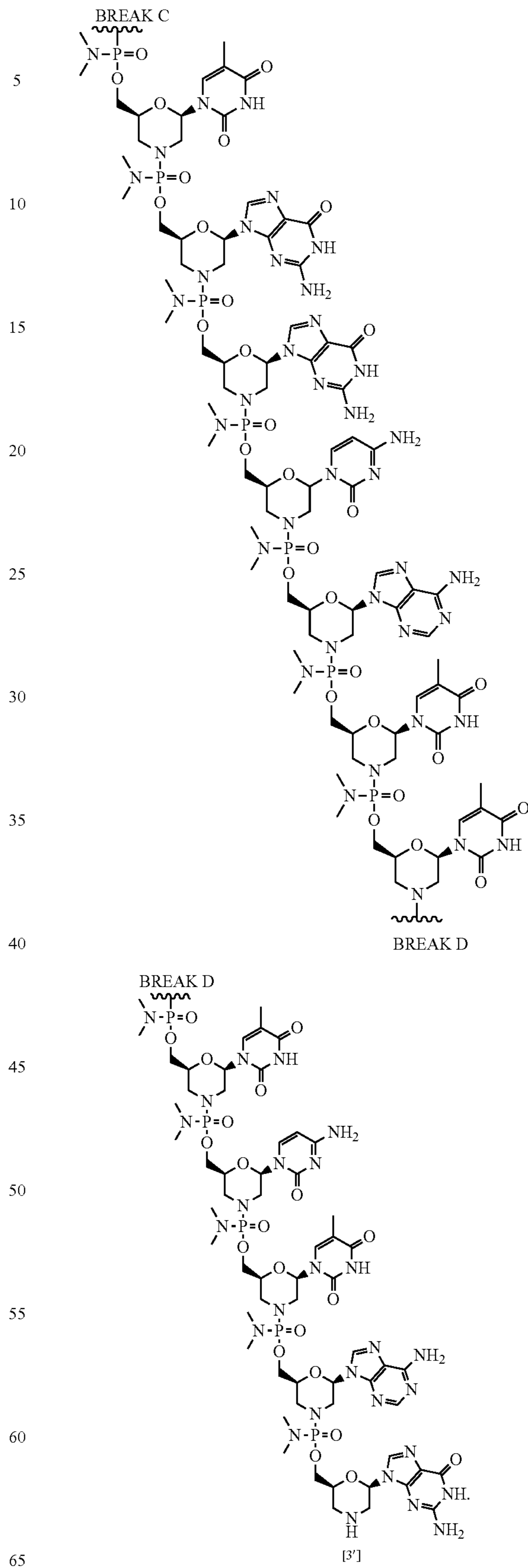
153

-continued

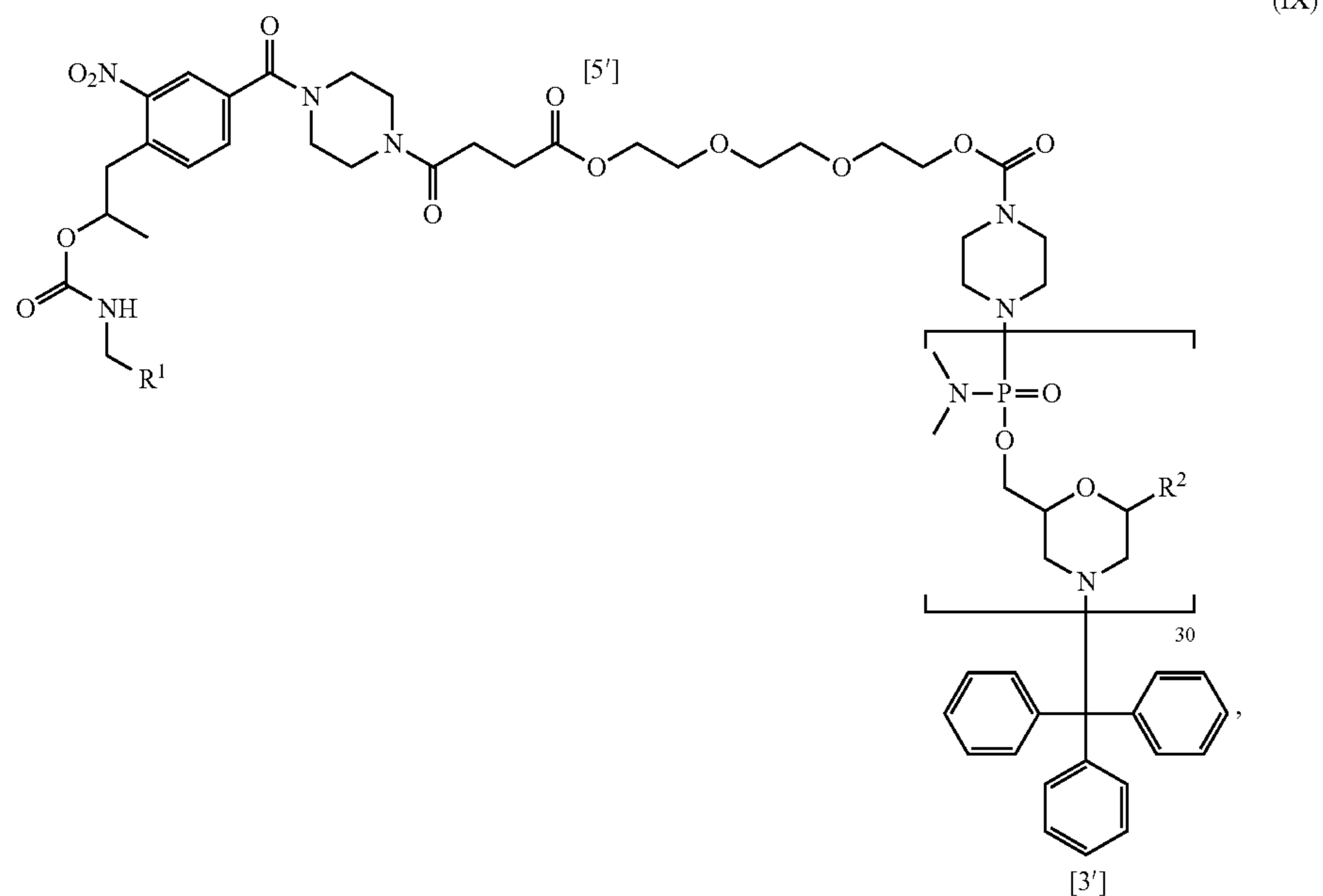


154

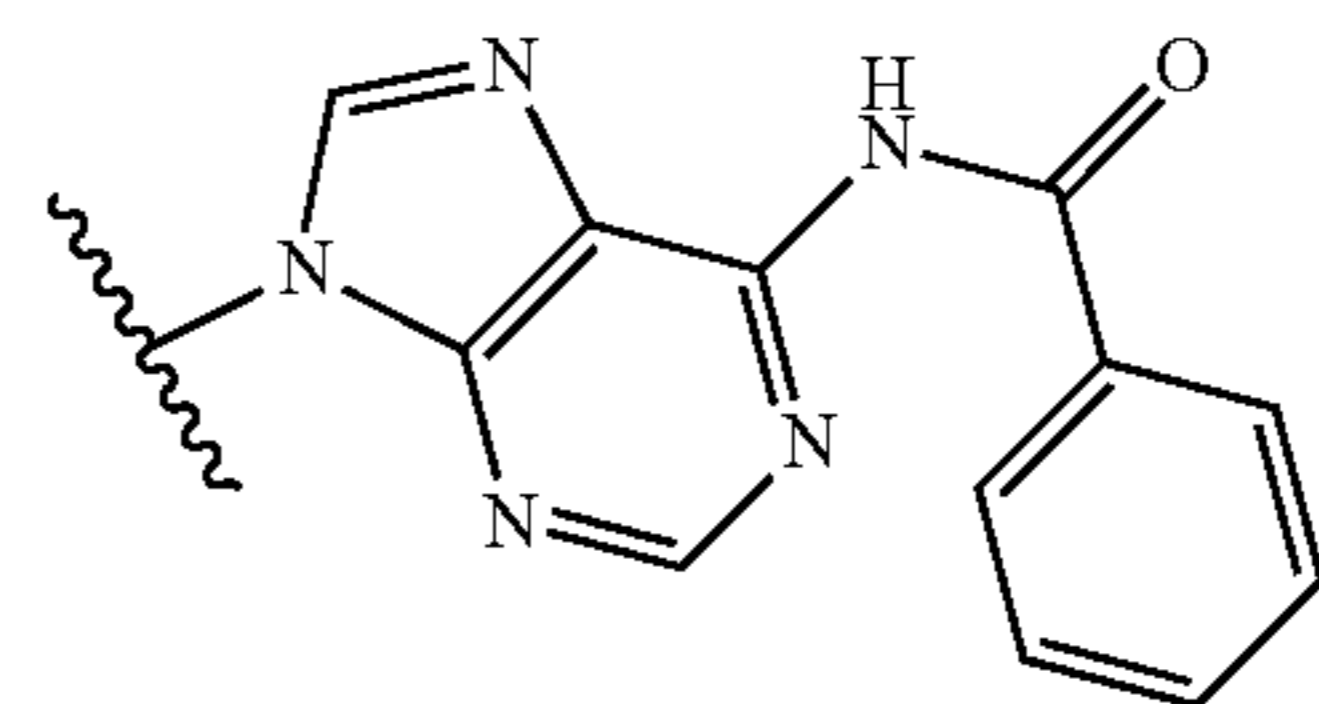
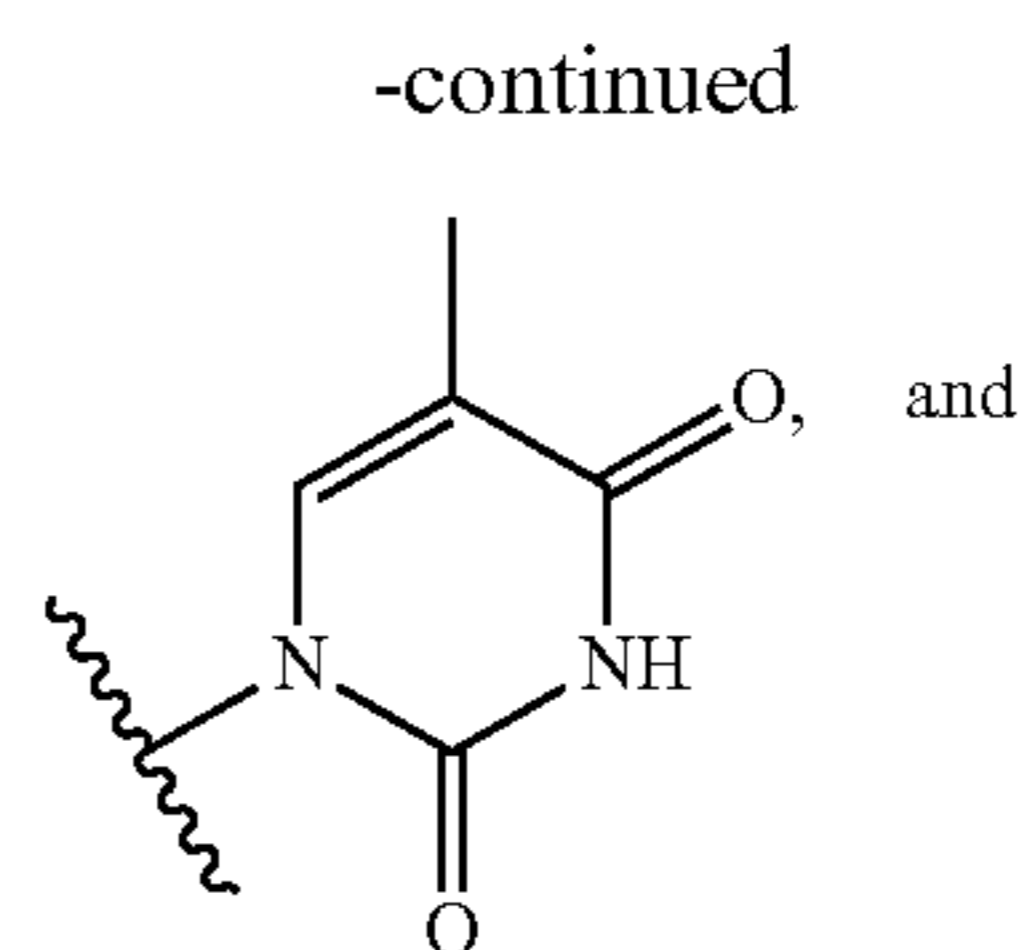
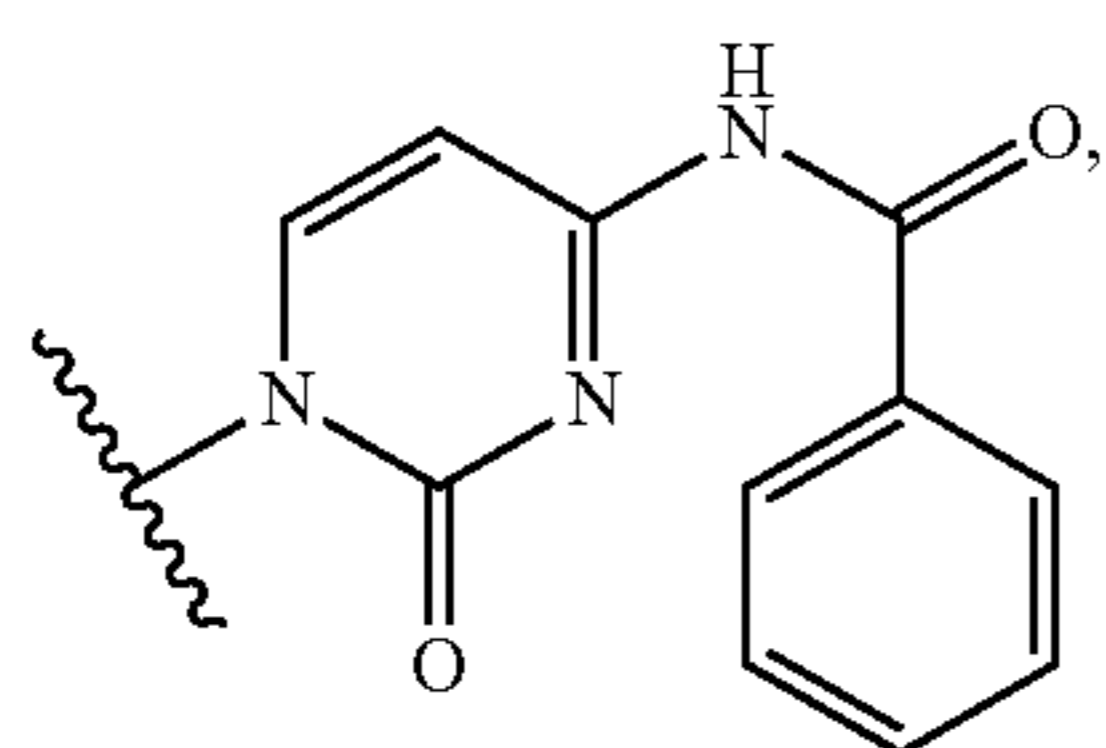
-continued



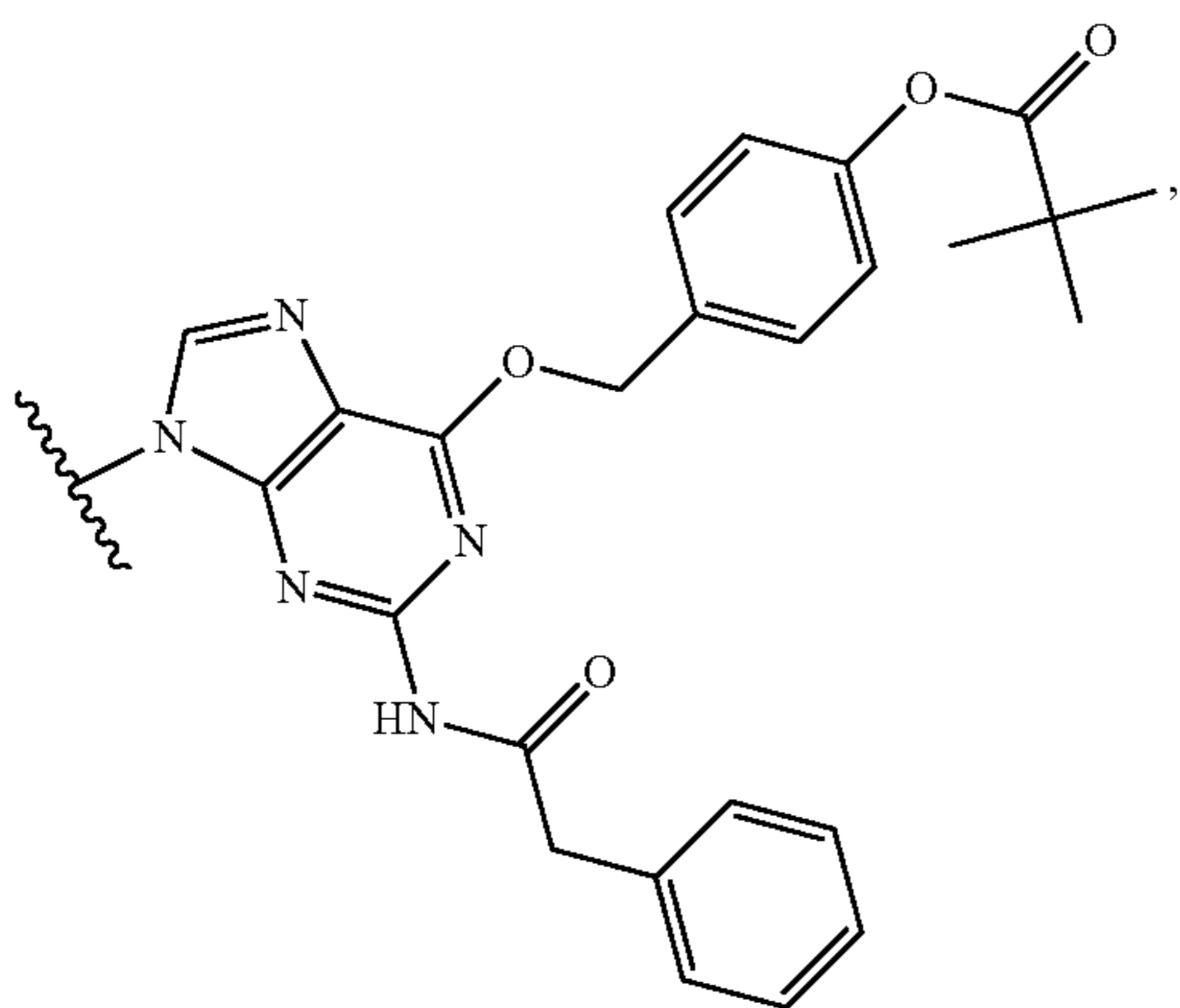
In another aspect, provided herein is a compound of Formula (IX):



or a pharmaceutically acceptable salt thereof, wherein:
 R^1 is a support-medium, and
 R^2 is, independently at each occurrence, selected from the group consisting of:



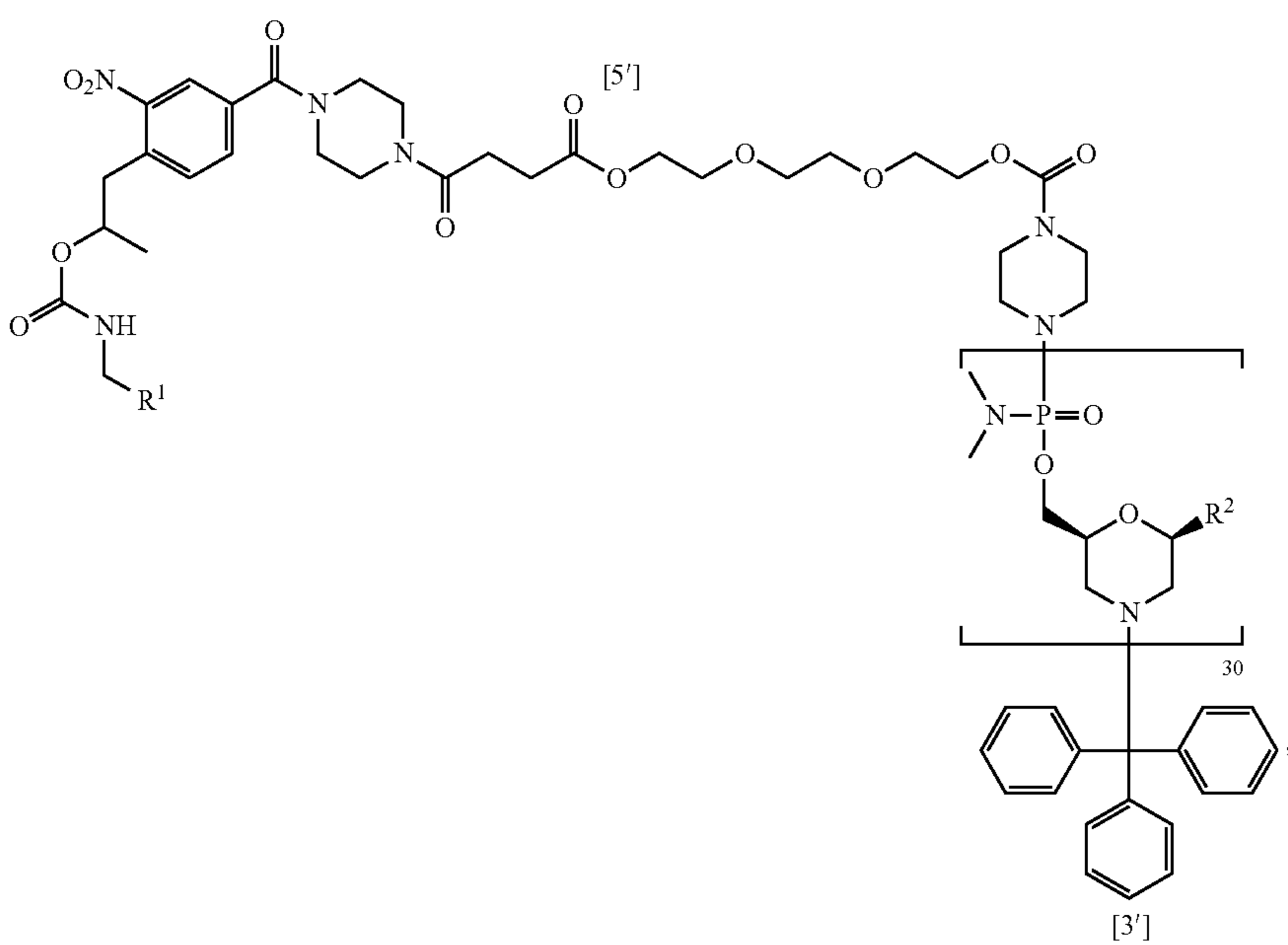
and
 wherein R^2 is at each position from 1 to 30 and 5' to 3':



Position No. 5' to 3'	R^2
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC
8	PA
9	T
10	PC
11	PA
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG

Position No. 5' to 3'	R ²	
18	PA	5
19	T	
20	DPG	
21	DPG	
22	PC	
23	PA	10
24	T	
25	T	
26	T	
27	PC	
28	T	15
29	PA	
30	DPG	

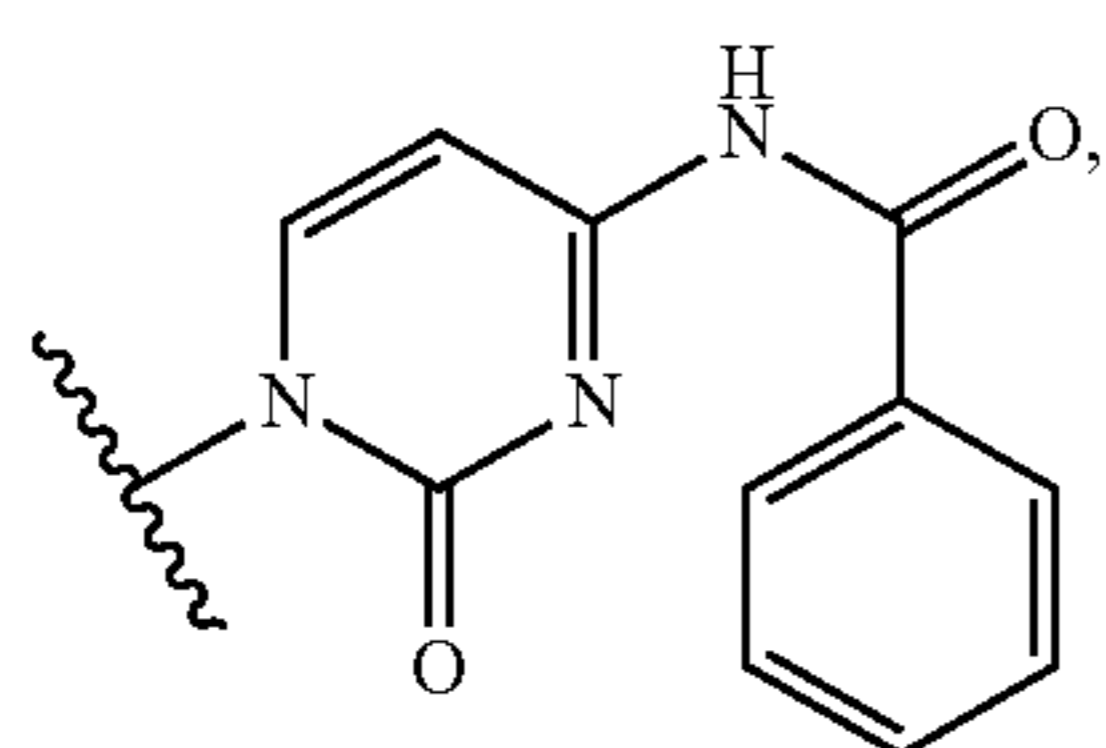
In one embodiment, the compound of Formula (IX) is of Formula (IXa):



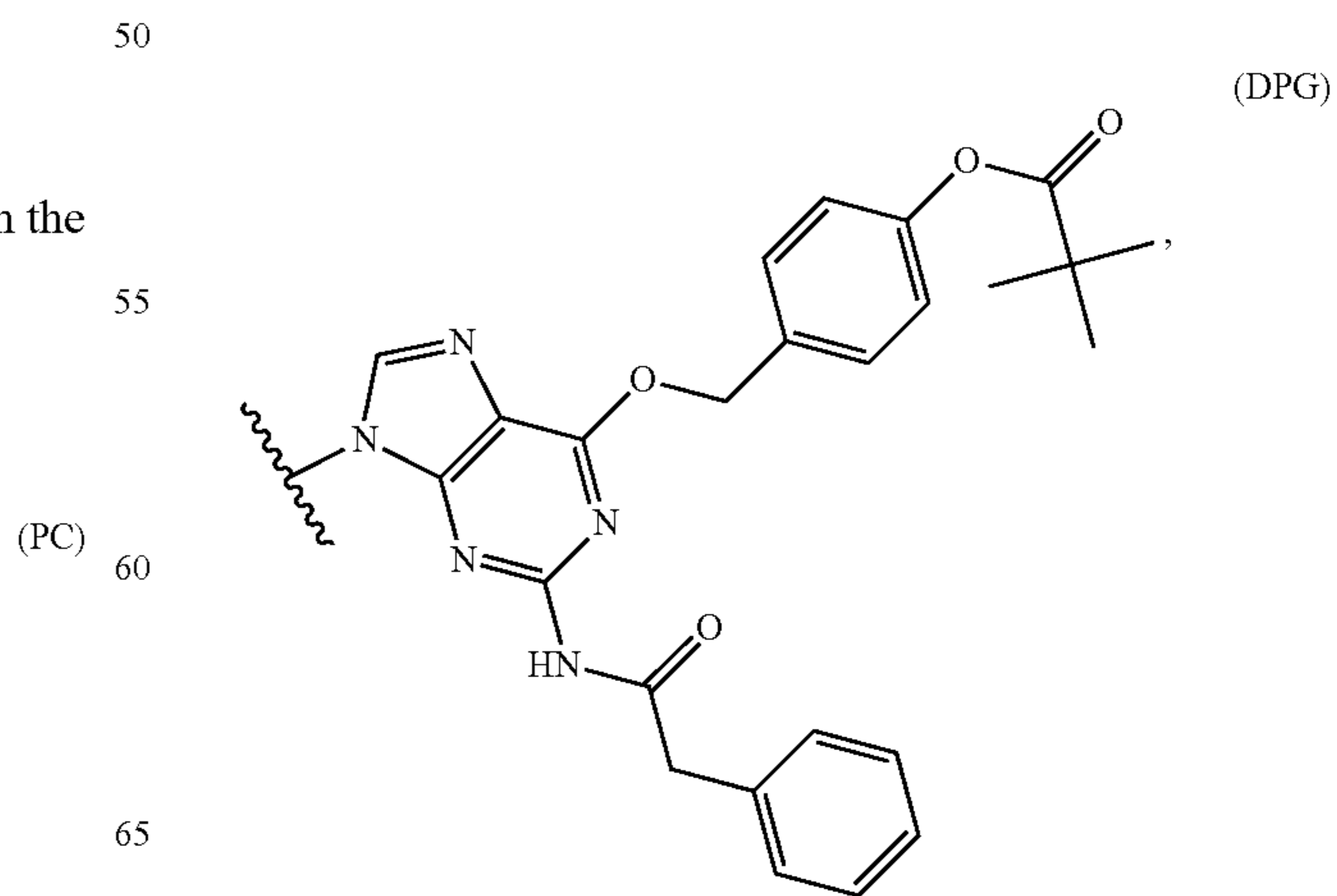
or a pharmaceutically acceptable salt thereof, wherein

R¹ is a support-medium, and

R² is, independently at each occurrence, selected from the group consisting of:

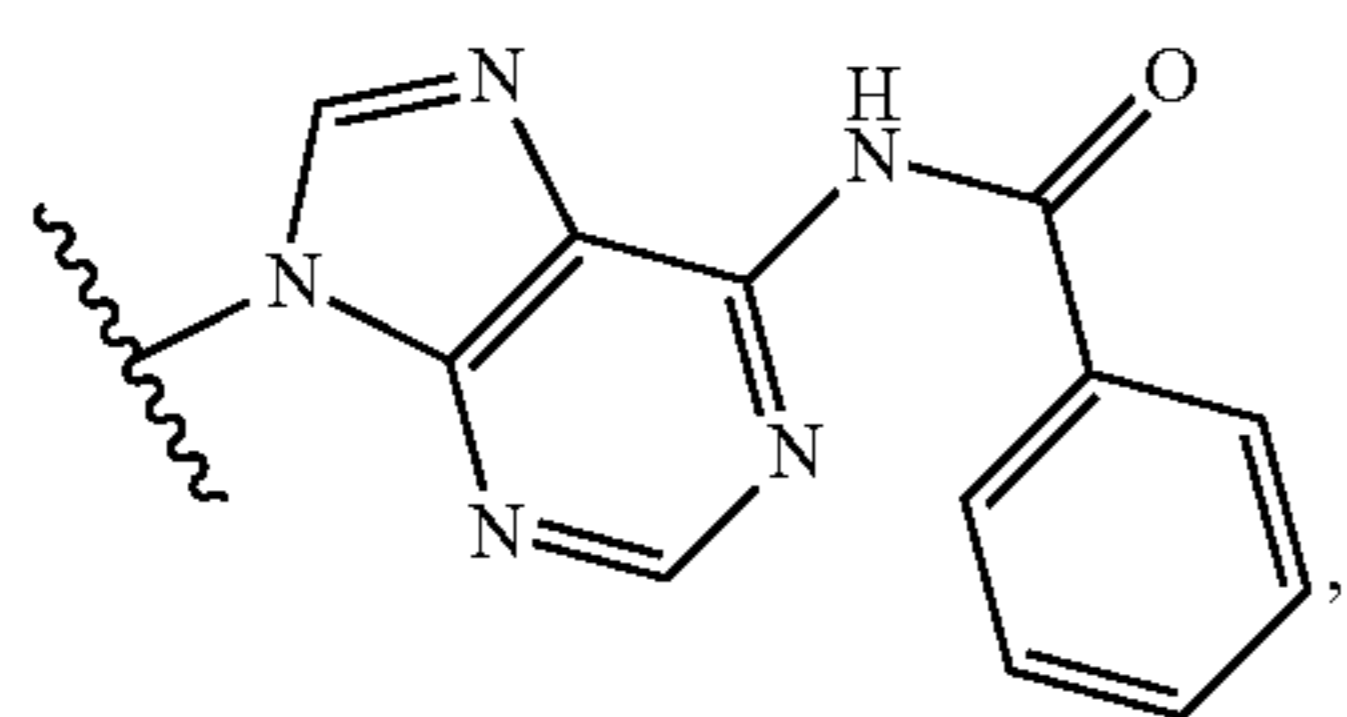
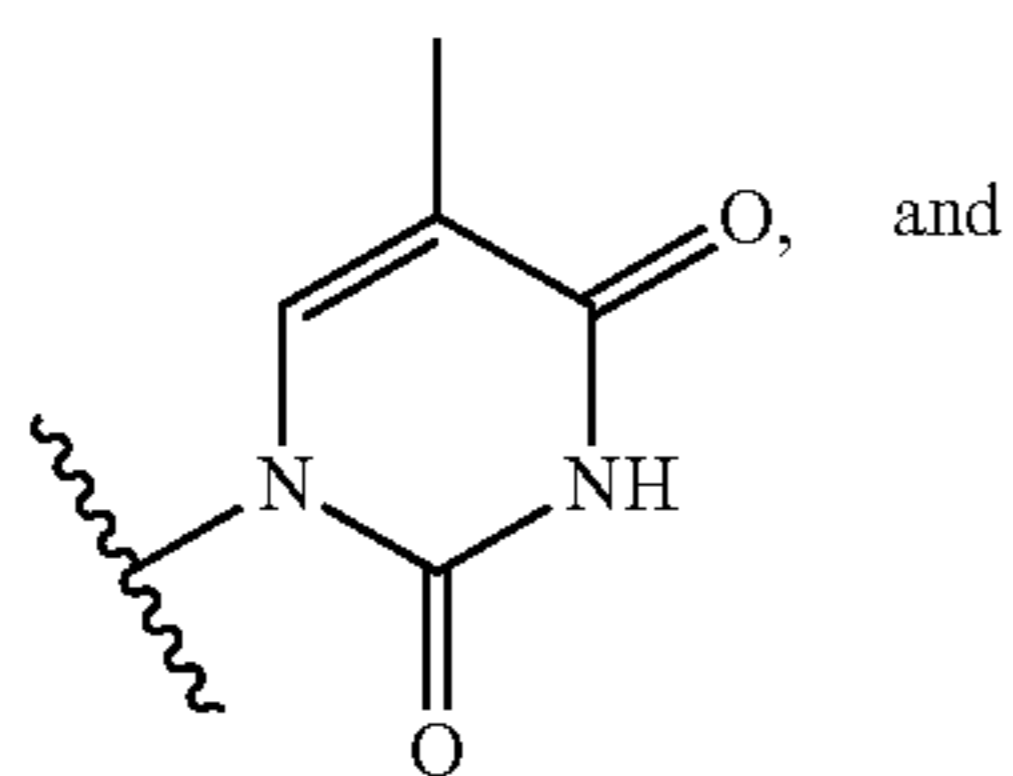


-continued



159

-continued



and

wherein R² is at each position from 1 to 30 and 5' to 3':

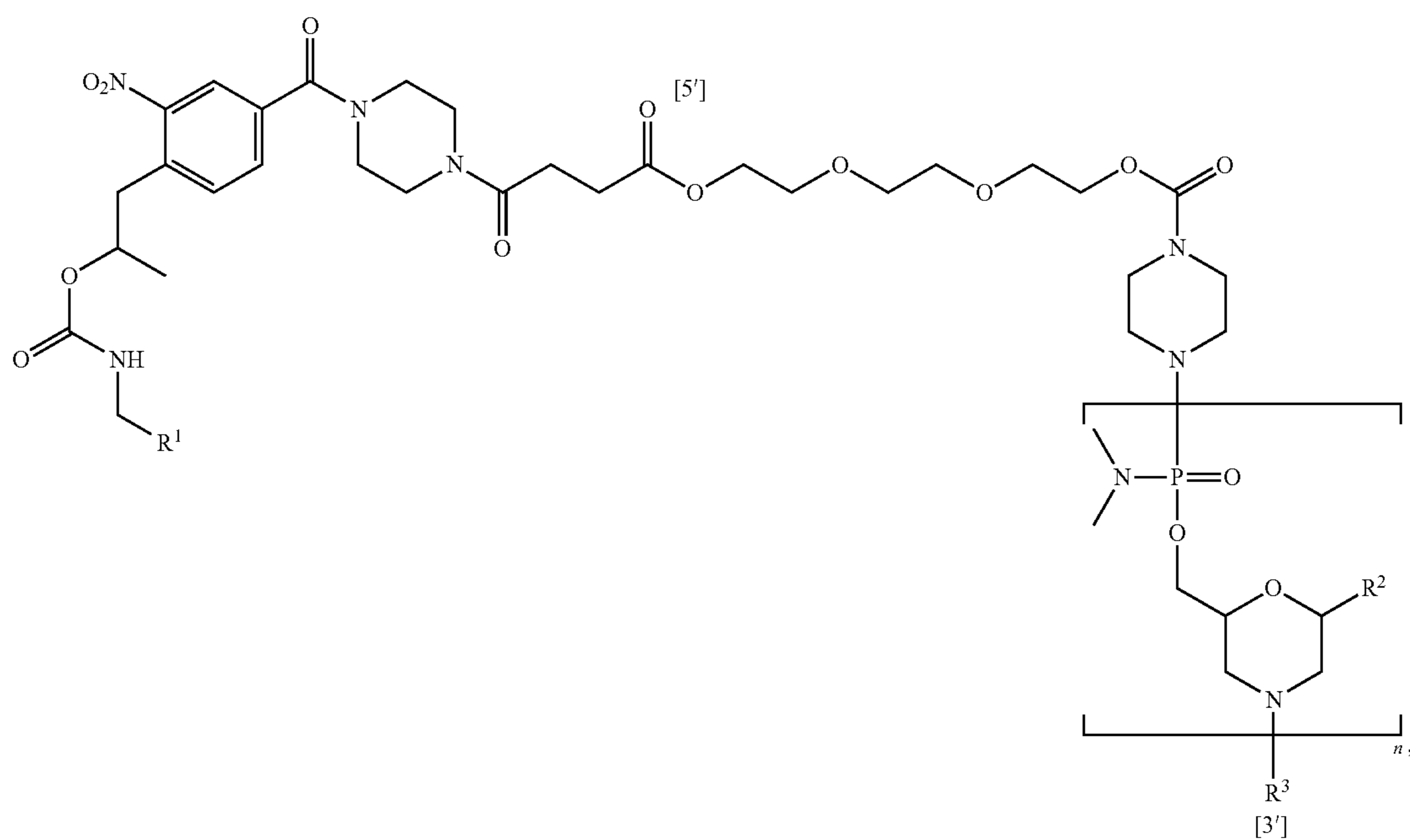
Position No. 5' to 3'	R ²
1	PC
2	T

160

-continued

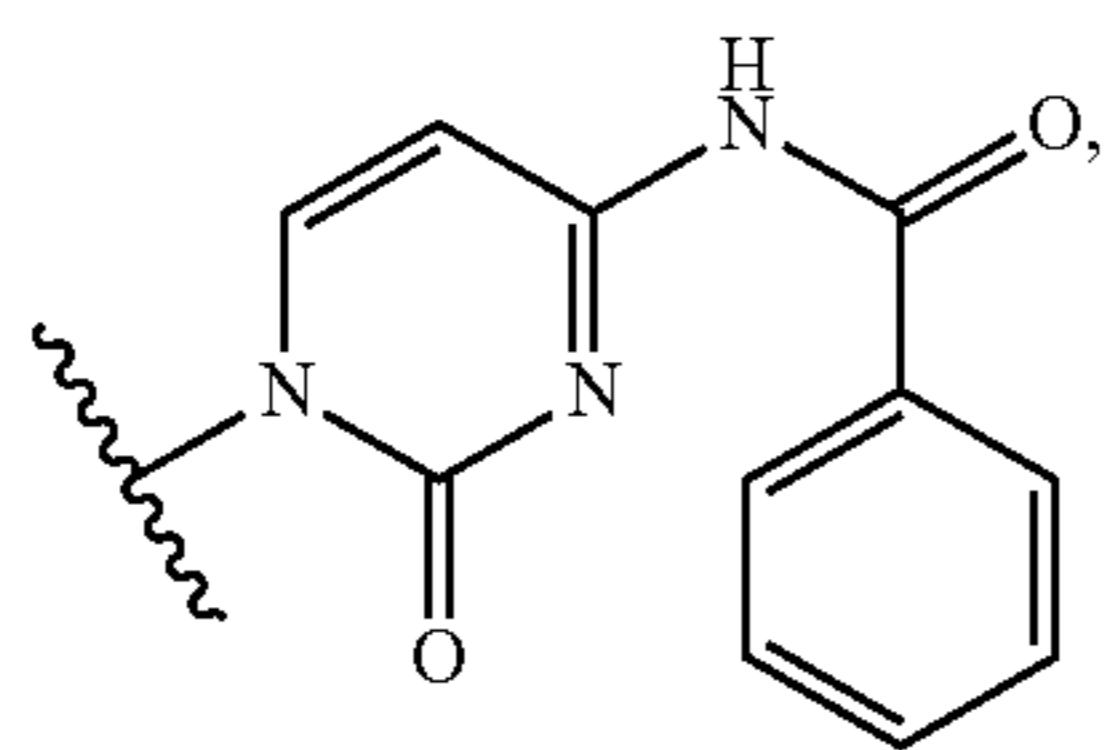
(T)	Position No. 5' to 3'	R ²
	3	PC
5	4	PC
	5	PA
	6	PA
	7	PC
	8	PA
	9	T
10	10	PC
	11	PA
	12	PA
	13	DPG
	14	DPG
	15	PA
(PA) 15	16	PA
	17	DPG
	18	PA
	19	T
	20	DPG
20	21	DPG
	22	PC
	23	PA
	24	T
	25	T
	26	T
	27	PC
	28	T
	29	PA
	30	DPG

30 In another aspect, provided herein is a compound of Formula (A9):



161

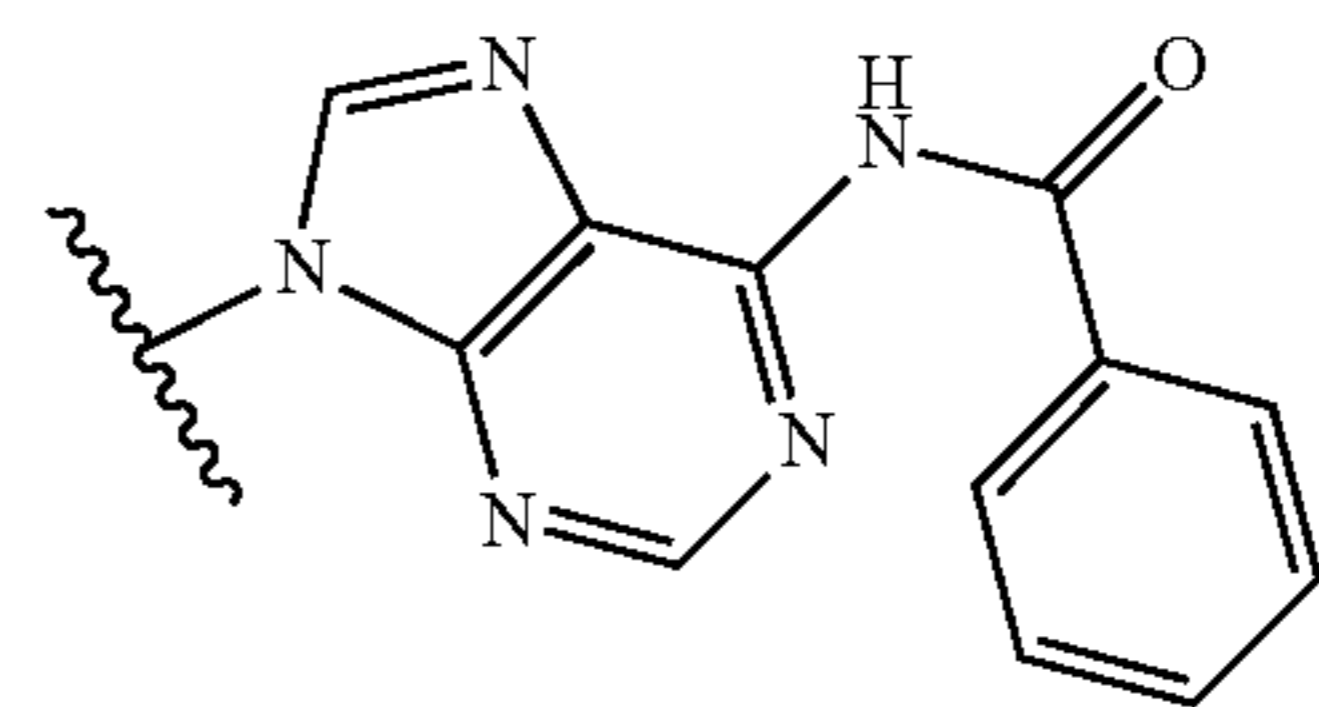
or a pharmaceutically acceptable salt thereof, wherein:
 n is an integer from 10 to 40;
 R¹ is a support-medium;
 R³ is selected from the group consisting of trityl,
 monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;
 and
 R⁴ is, independently at each occurrence, selected from the
 group consisting of:



(PC)

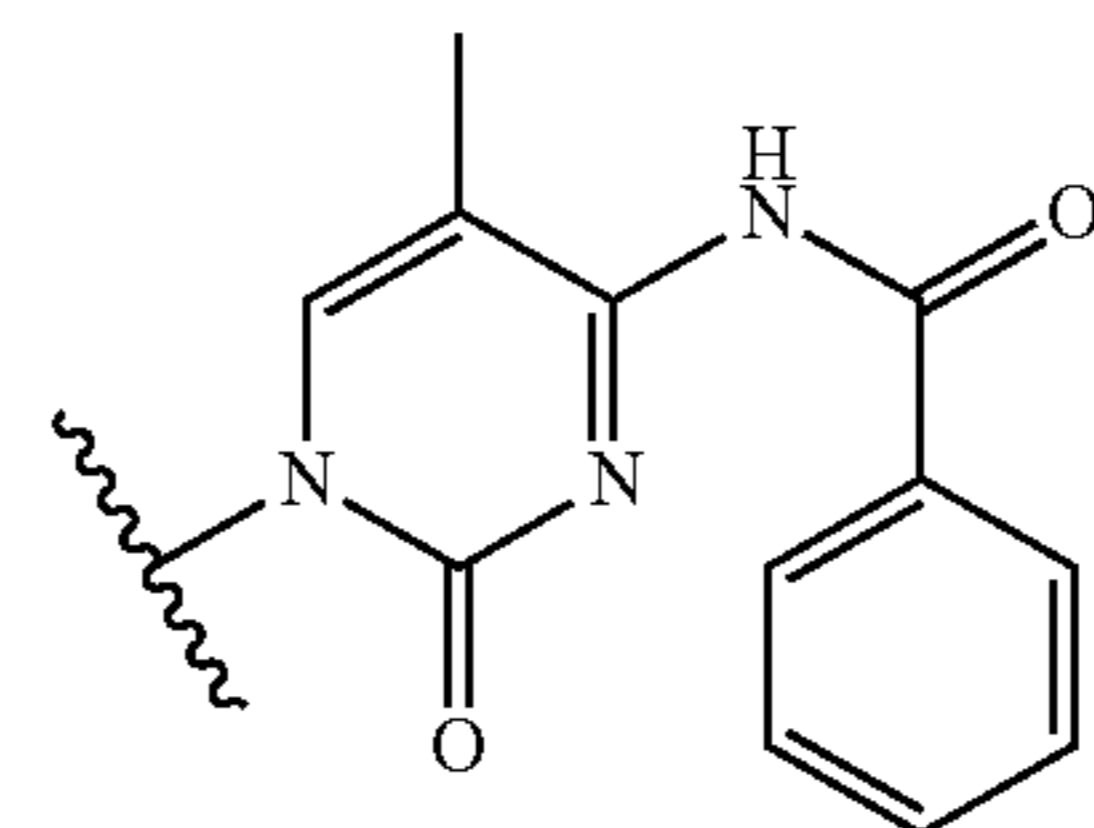
162

-continued



(PA)

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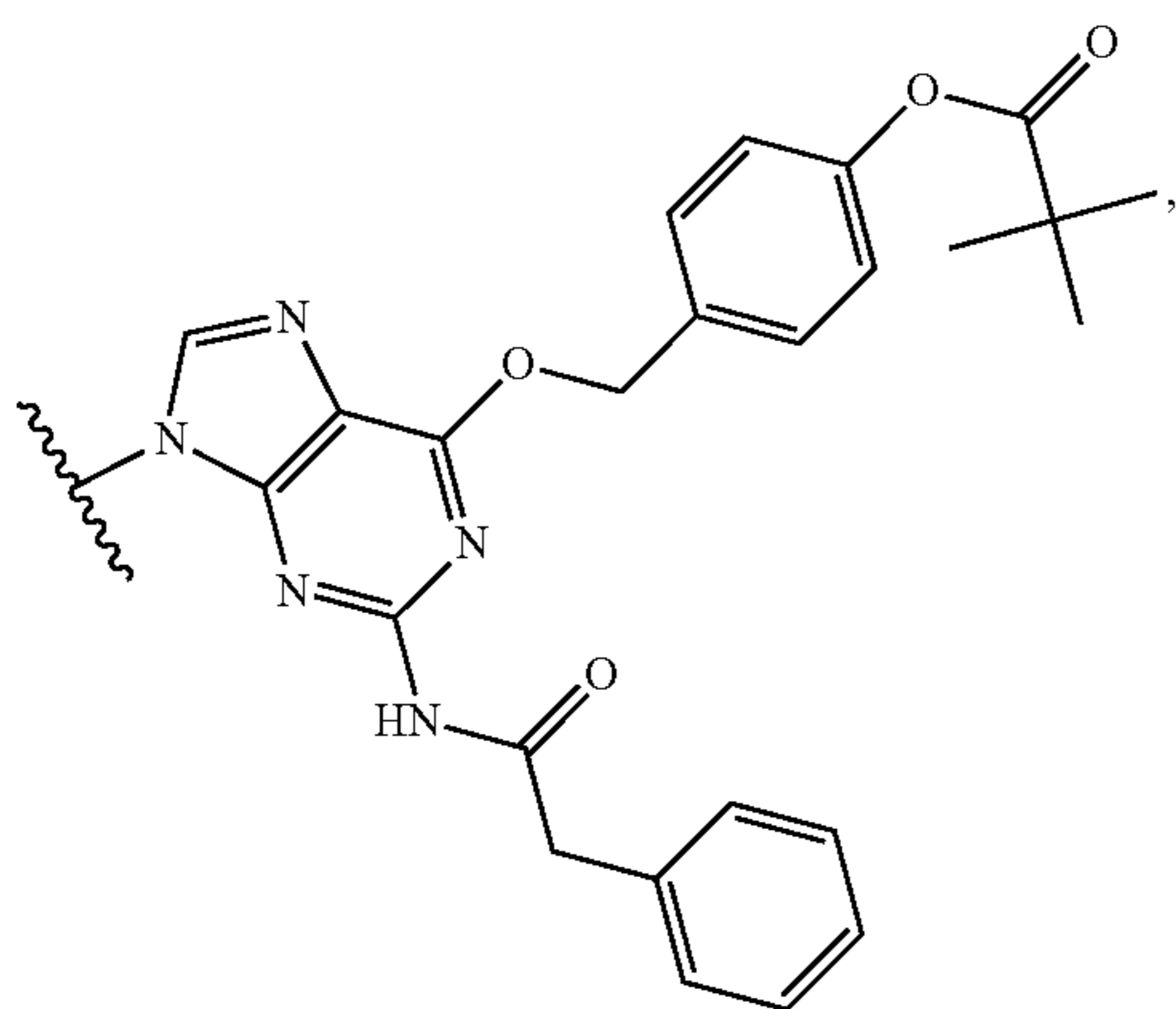


(P5mC)

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(DPG)

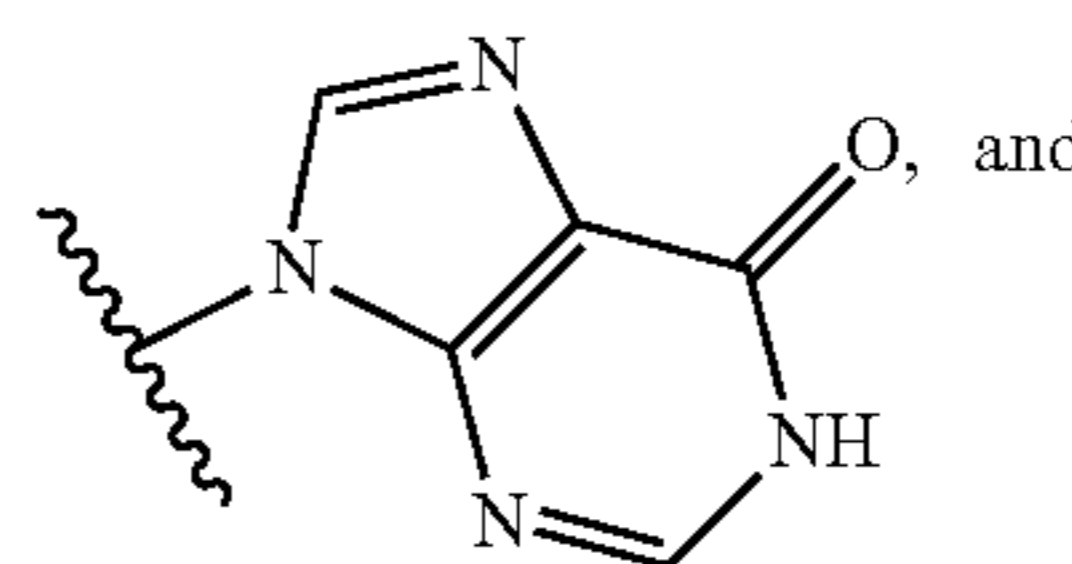
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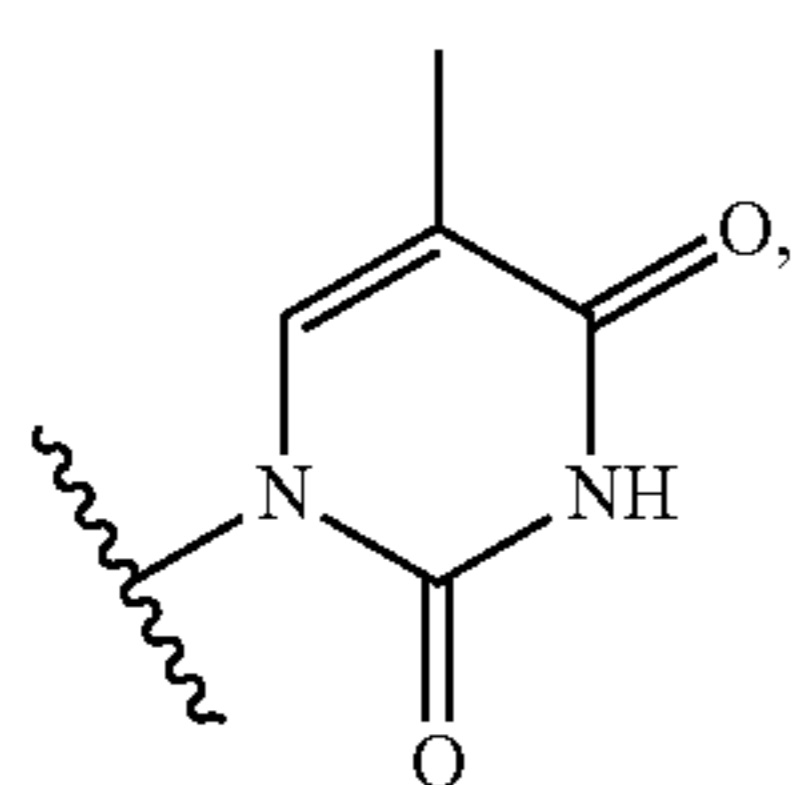


(U)

(I)

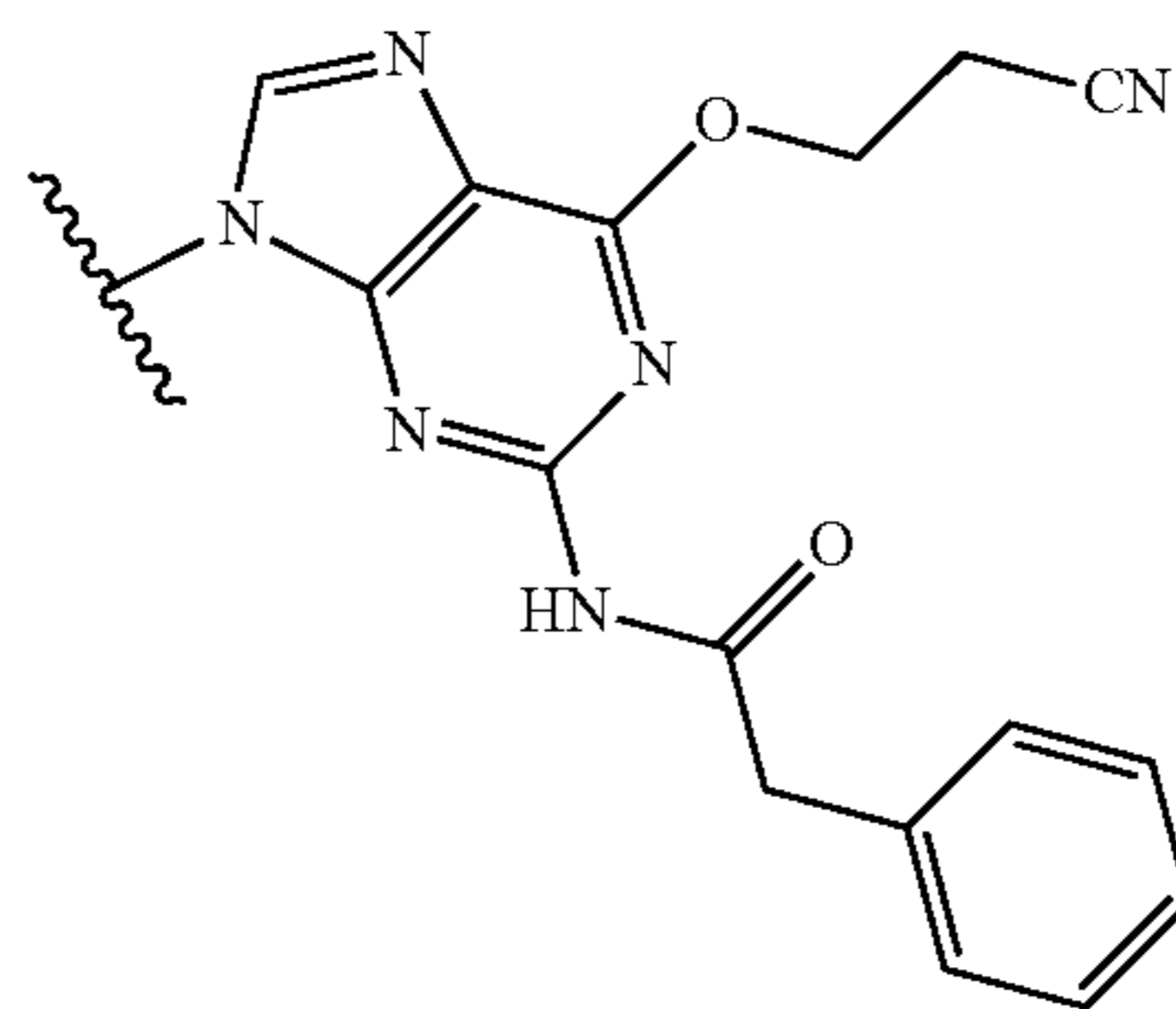
55

(T)



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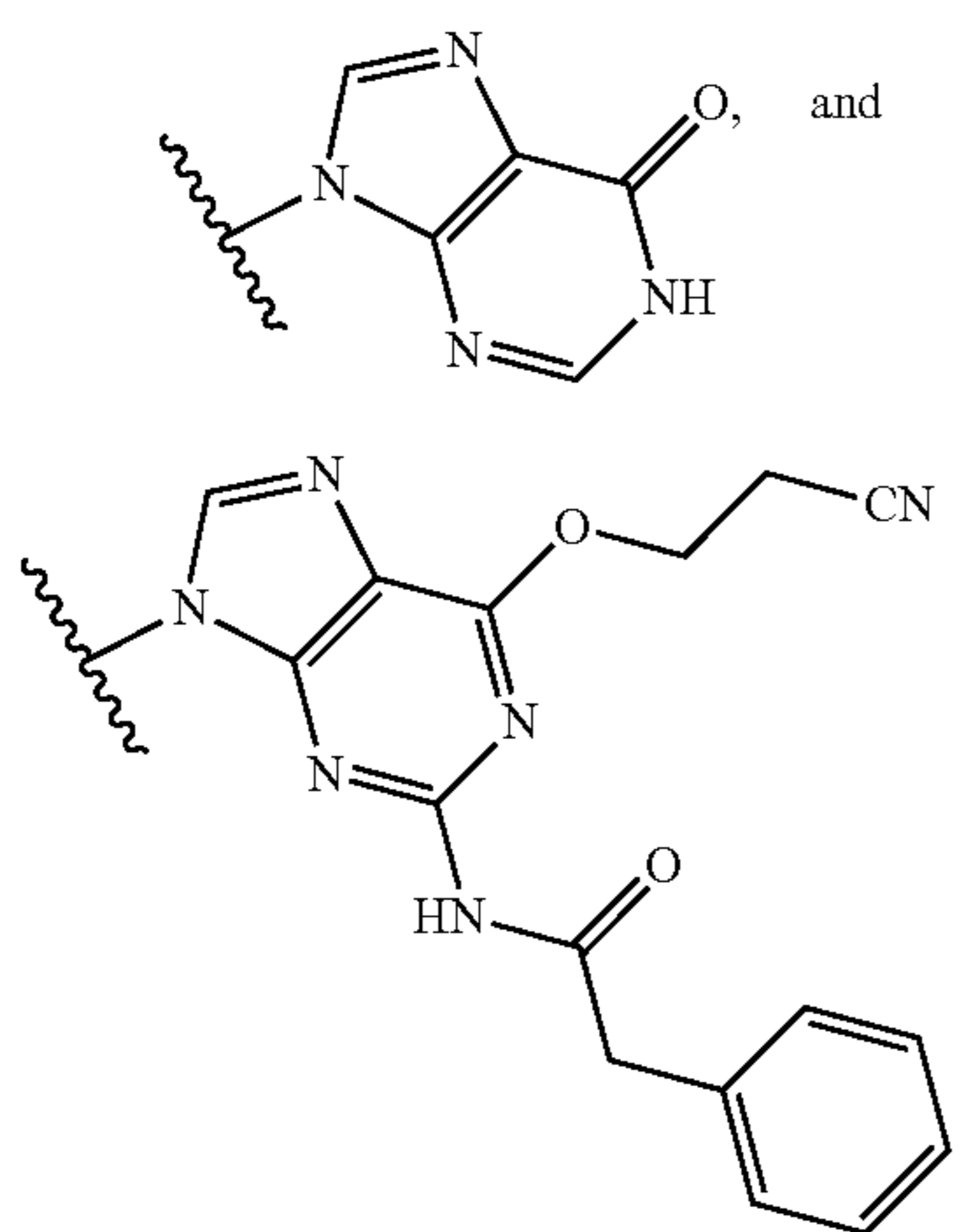
65



(PG)

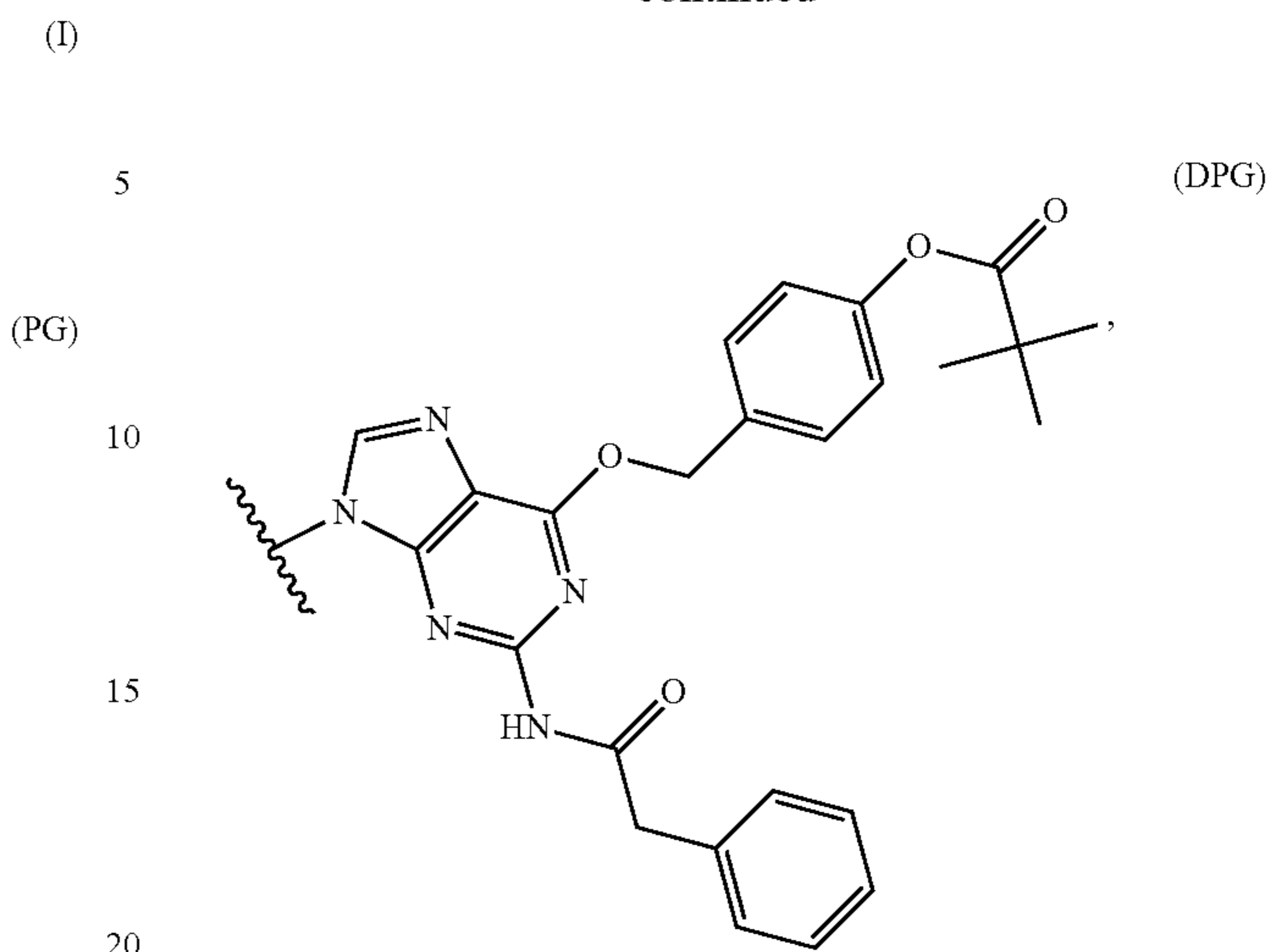
165

-continued

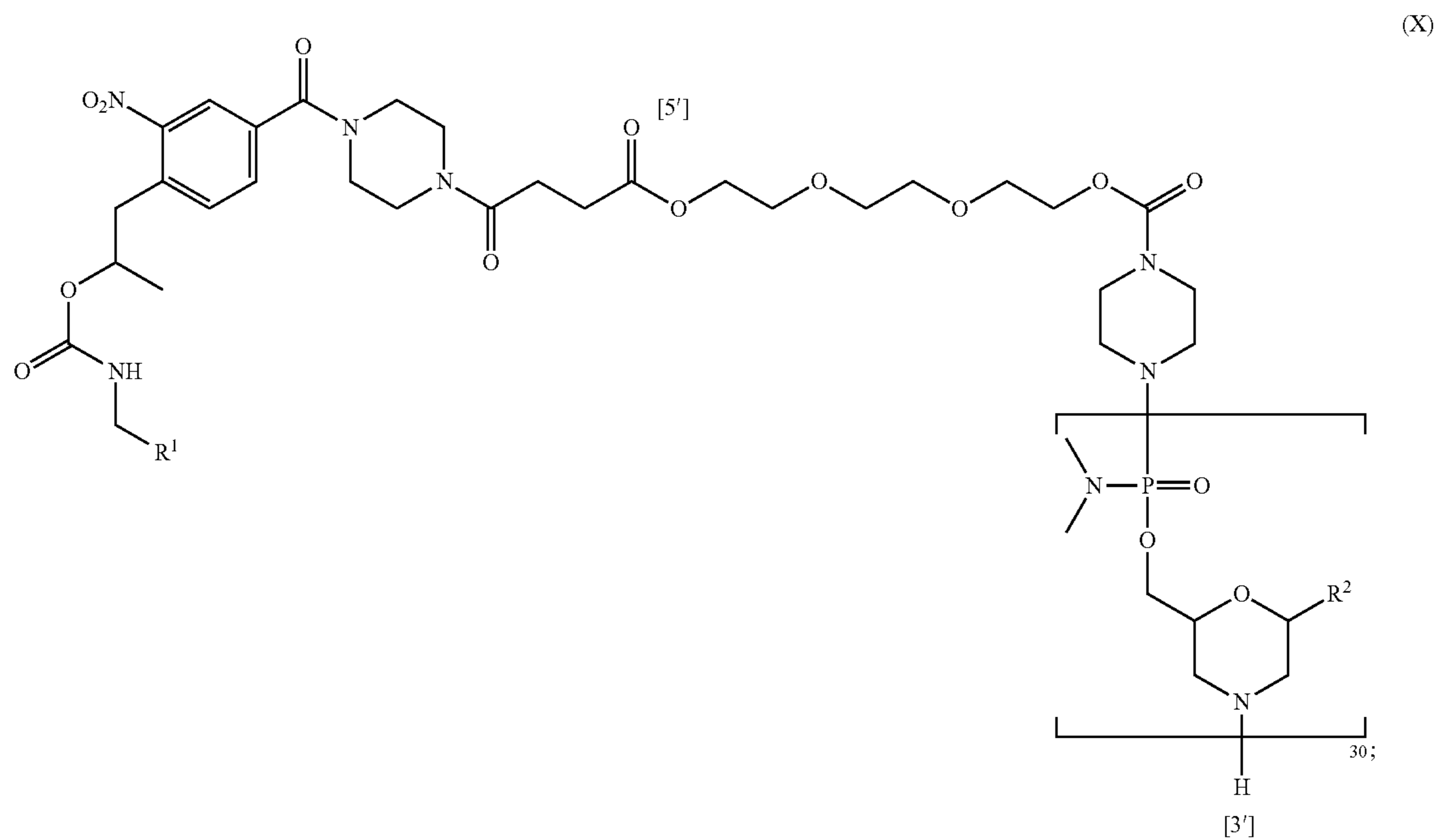


166

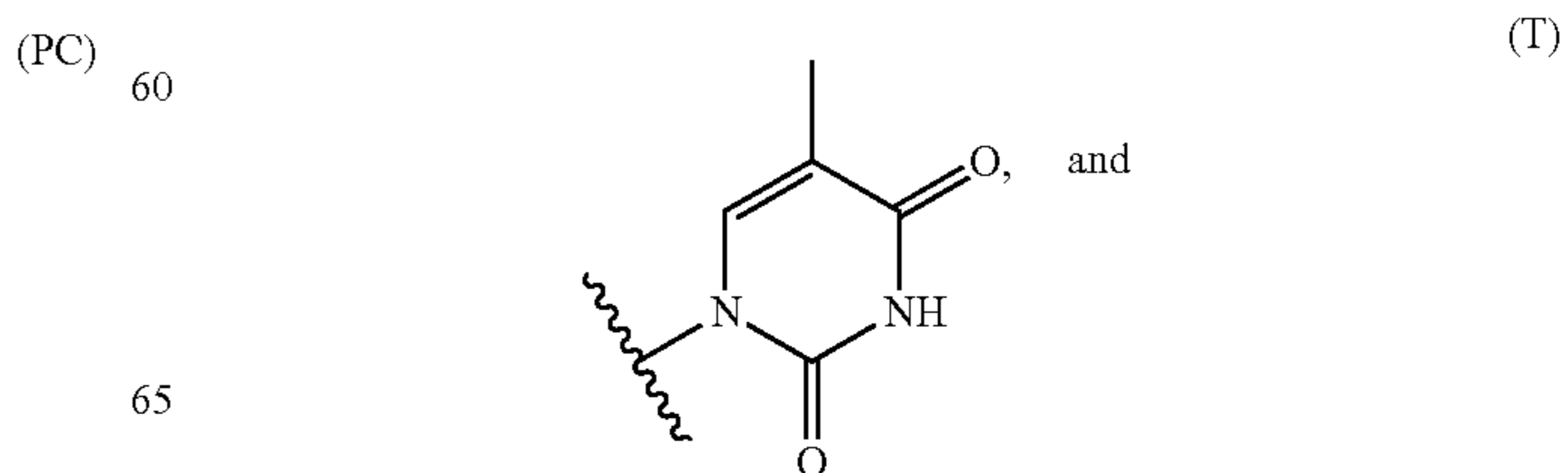
-continued



In another aspect, provided herein is a compound of Formula (X):

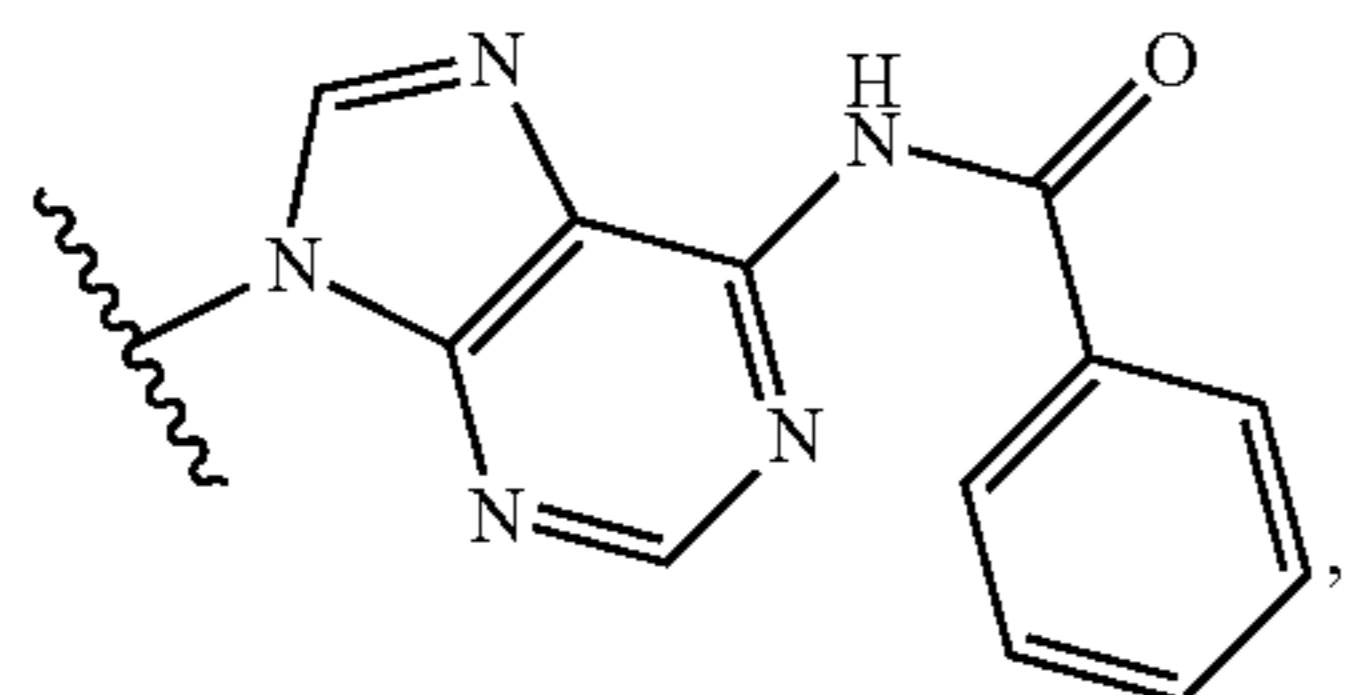


or a pharmaceutically acceptable salt thereof, wherein R^1 is a support-medium, and R^2 is, independently at each occurrence, selected from the group consisting of:



167

-continued



and

wherein R² is at each position from 1 to 30 and 5' to 3':

Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC

168

-continued

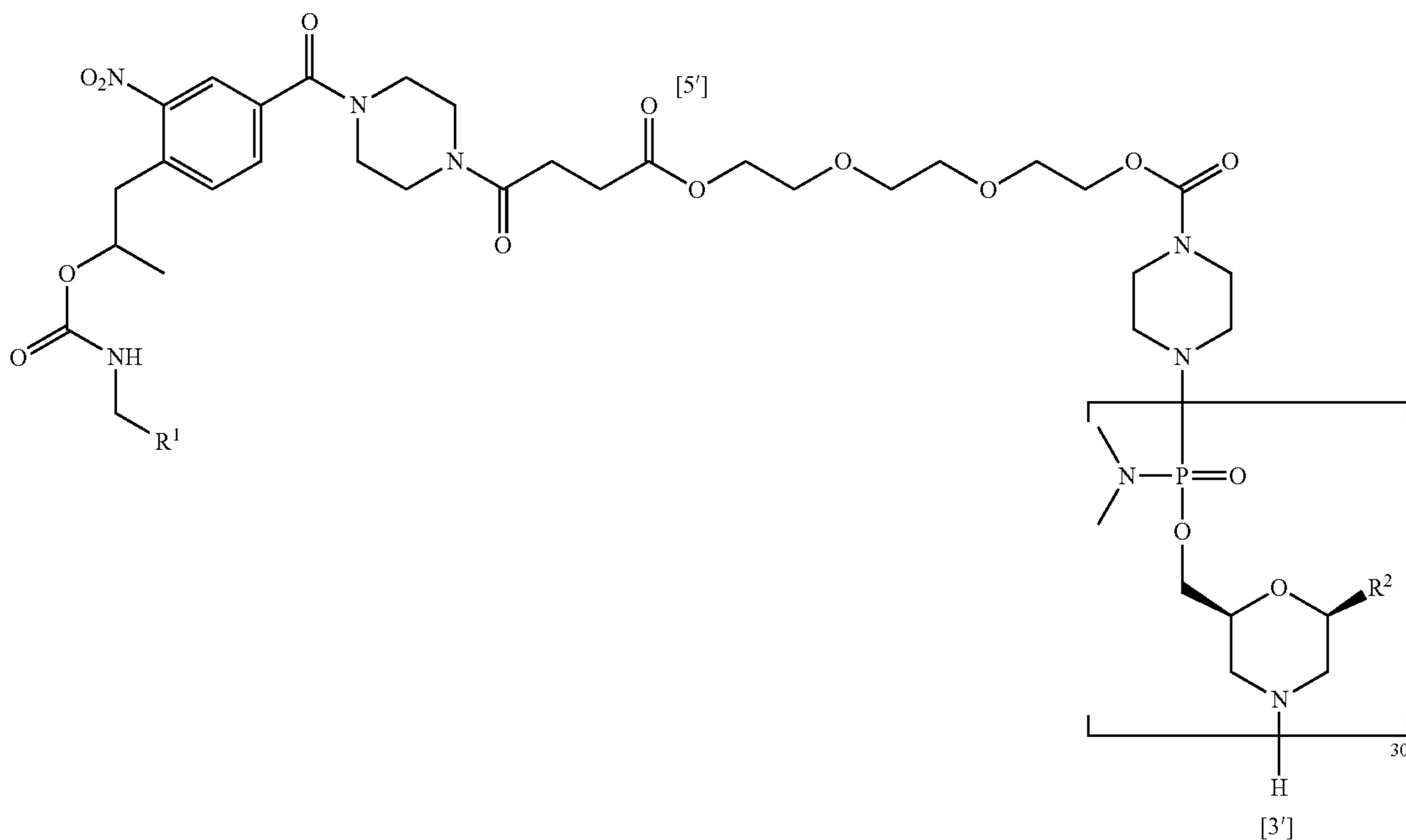
(PA)

Position No. 5' to 3'	R ²	
5	22	PC
	23	PA
	24	T
	25	T
10	26	T
	27	PC
	28	T
	29	PA
15	30	DPG

20

In one embodiment, the compound of Formula (X) is of Formula (Xa):

(Xa)



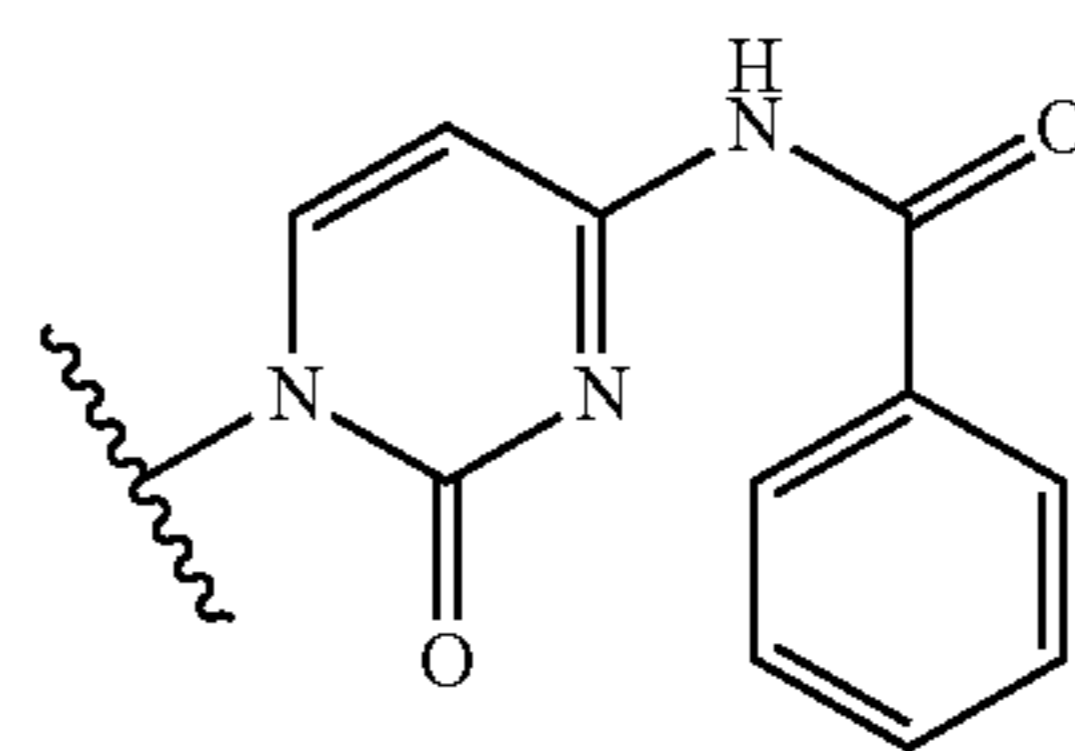
-continued

Position No. 5' to 3'	R ²
8	PA
9	T
10	PC
11	PA
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG
18	PA
19	T
20	DPG
21	DPG

or a pharmaceutically acceptable salt thereof, wherein R¹ is a support-medium, and

55 R² is, independently at each occurrence, selected from the group consisting of:

60

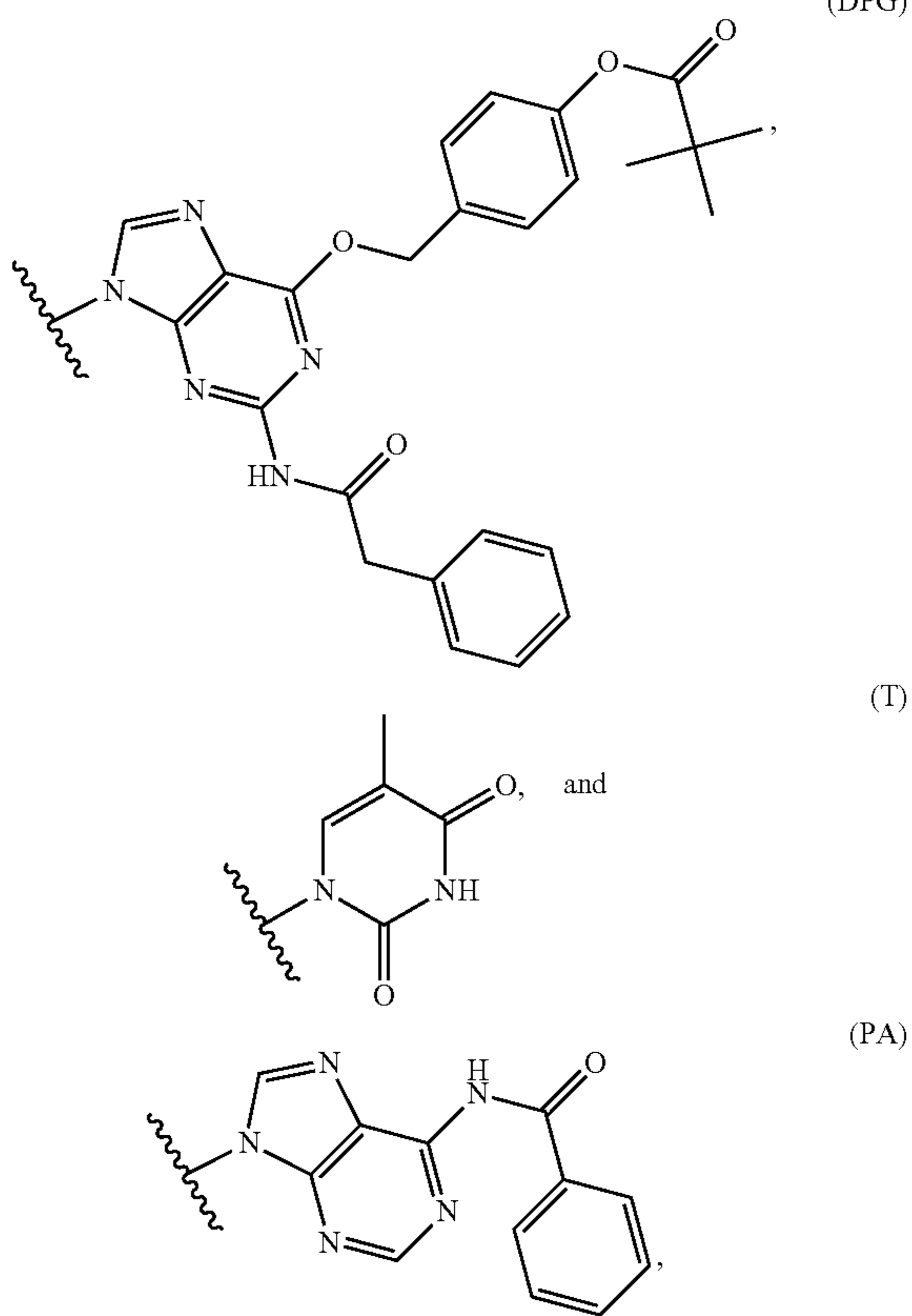


65

(PC)

169

-continued



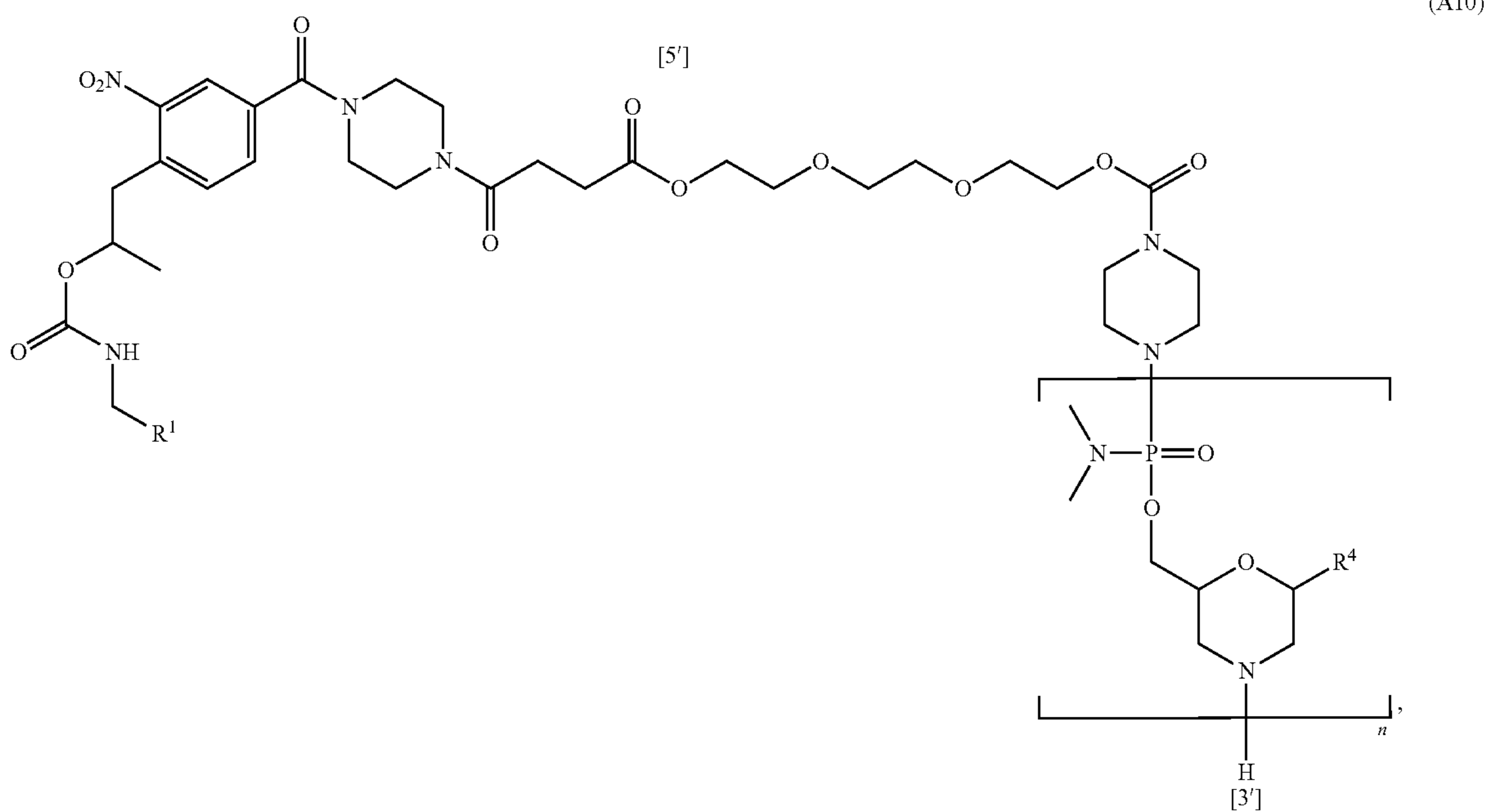
170

and

wherein R² is at each position from 1 to 30 and 5' to 3':

	Position No. 5' to 3'	R ²
5	1	PC
	2	T
	3	PC
10	4	PC
	5	PA
	6	PA
15	7	PC
	8	PA
	9	T
20	10	PC
	11	PA
	12	PA
(T)	13	DPG
	14	DPG
	15	PA
25	16	PA
	17	DPG
	18	PA
(PA)	19	T
	20	DPG
	21	DPG
30	22	PC
	23	PA
	24	T
	25	T
	26	T
	27	PC
	28	T
	29	PA
	30	DPG

In another aspect, provided herein is a compound of Formula (A10):



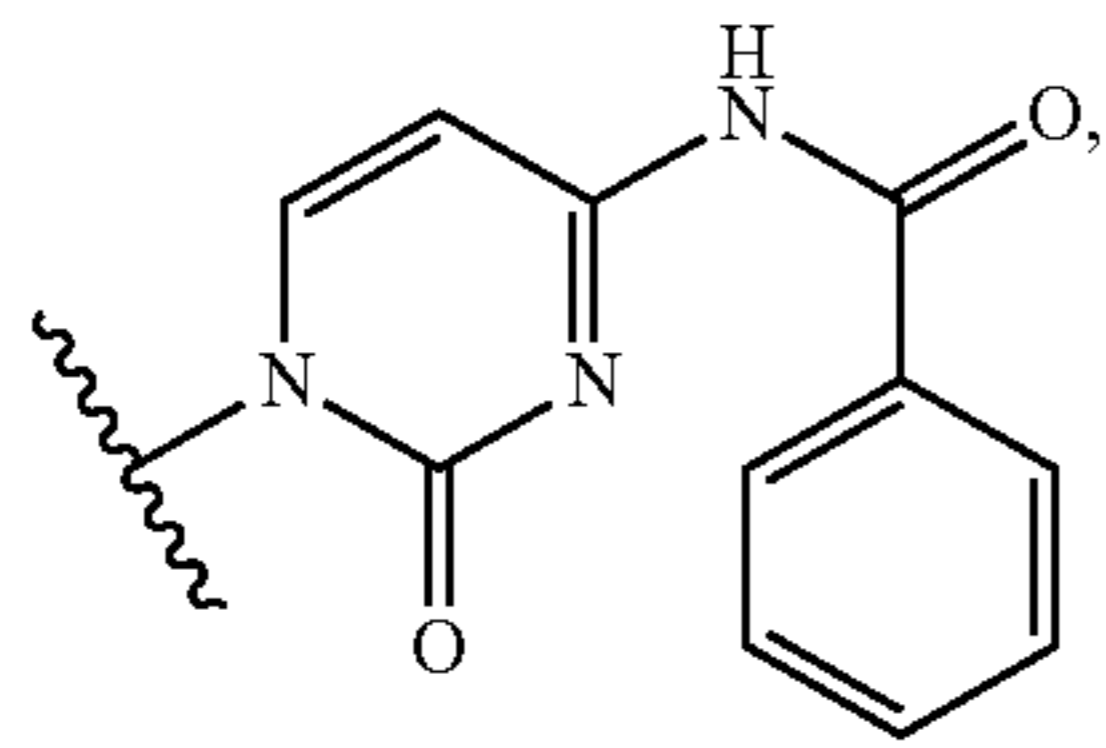
171

or a pharmaceutically acceptable salt thereof, wherein:

n is an integer from 10 to 40;

R¹ is a support-medium; and

R⁴ is, independently at each occurrence, selected from the group consisting of:



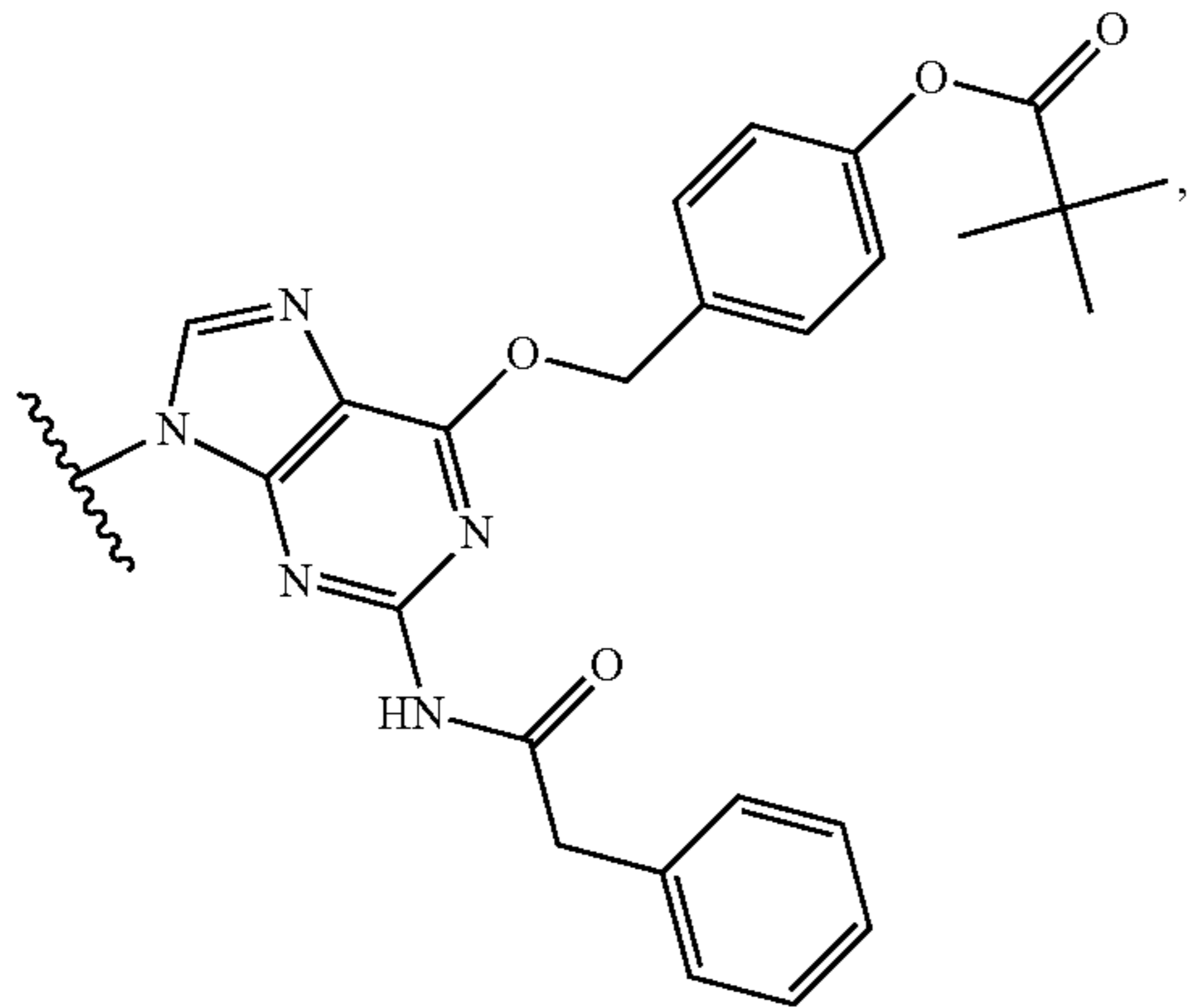
(PC)

10

15

20

(DPG)



25

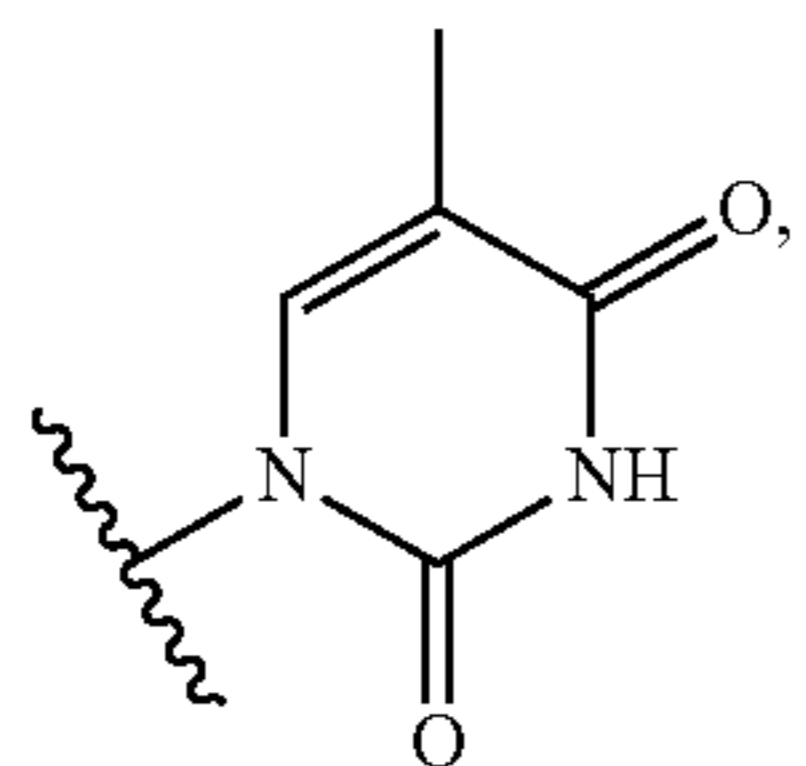
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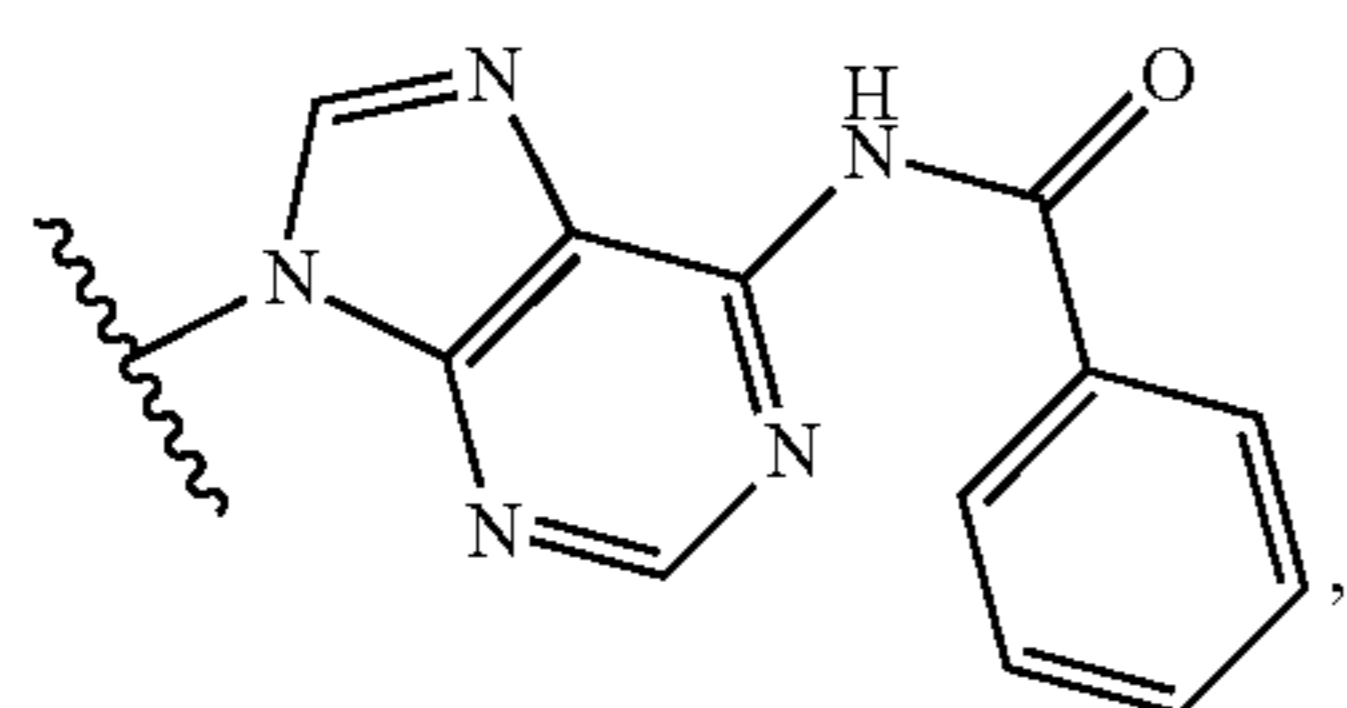
(T)

45



50

and



(PA)

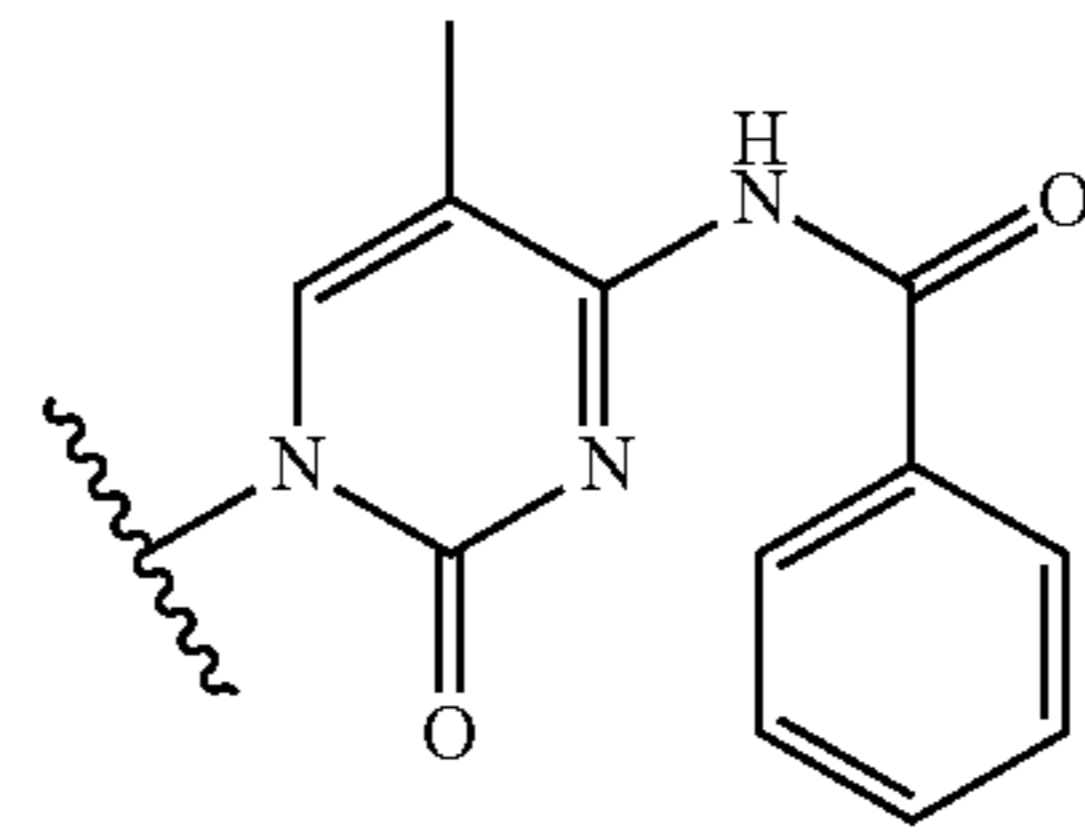
60

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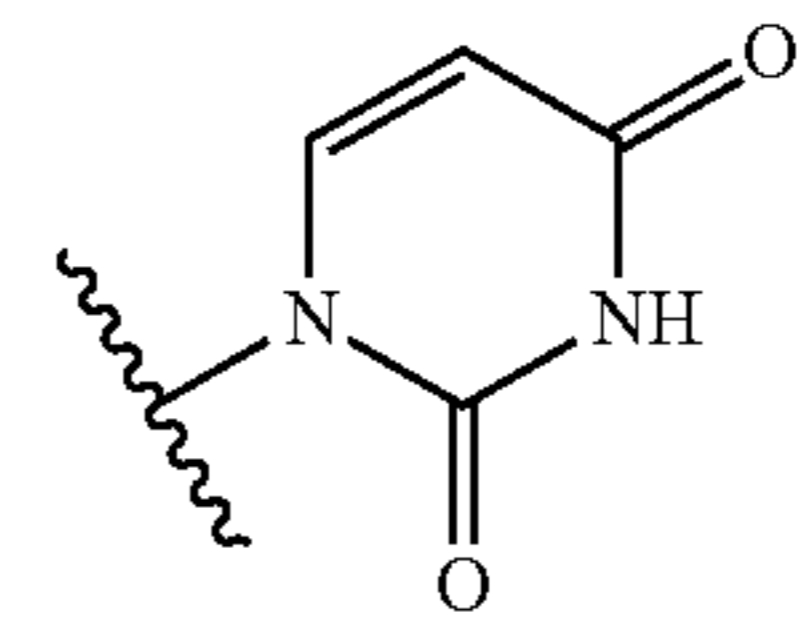
172

-continued

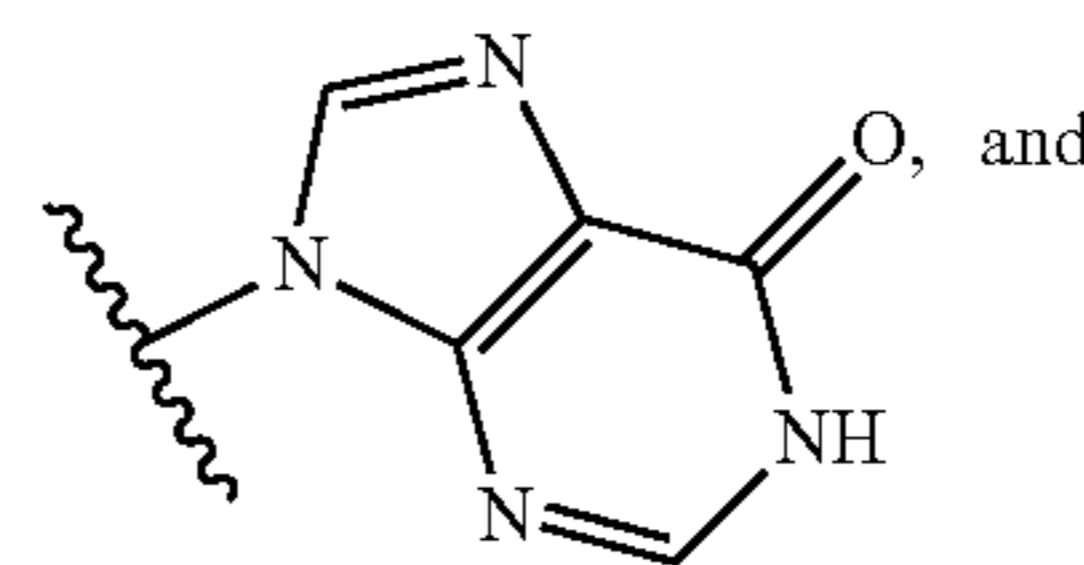
(P5mC)



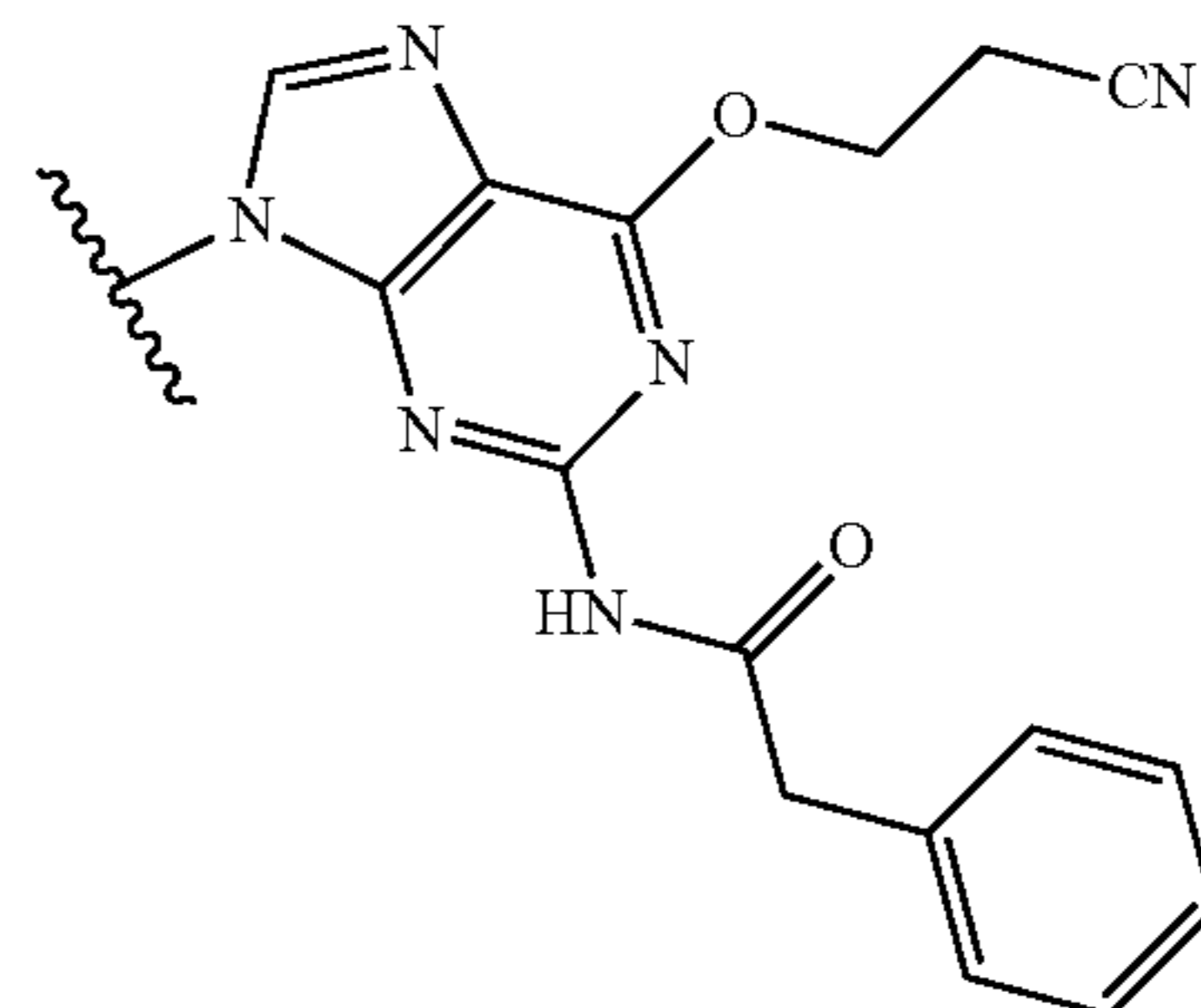
(U)



(I)



(PG)



45

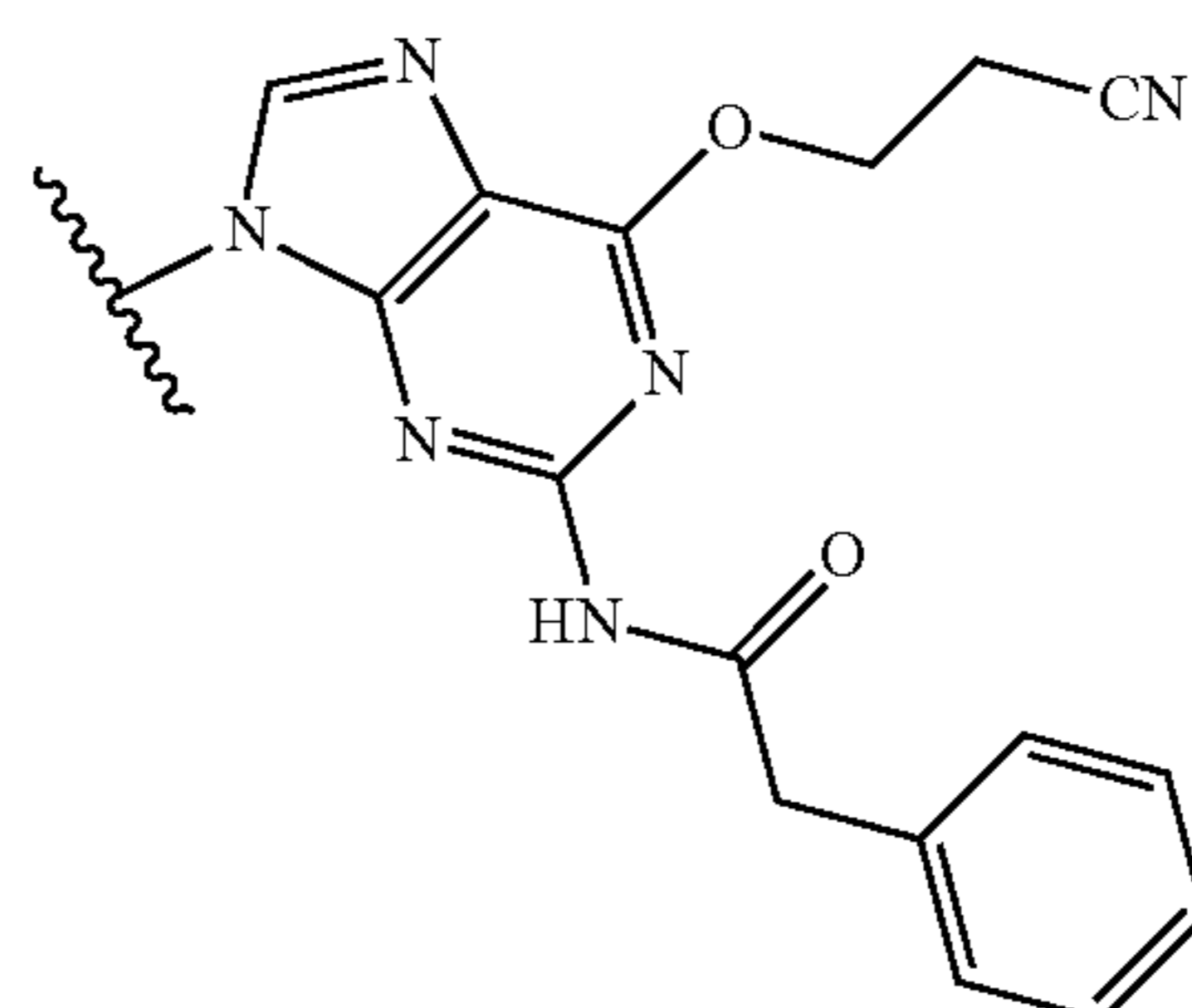
50

55

(PA)

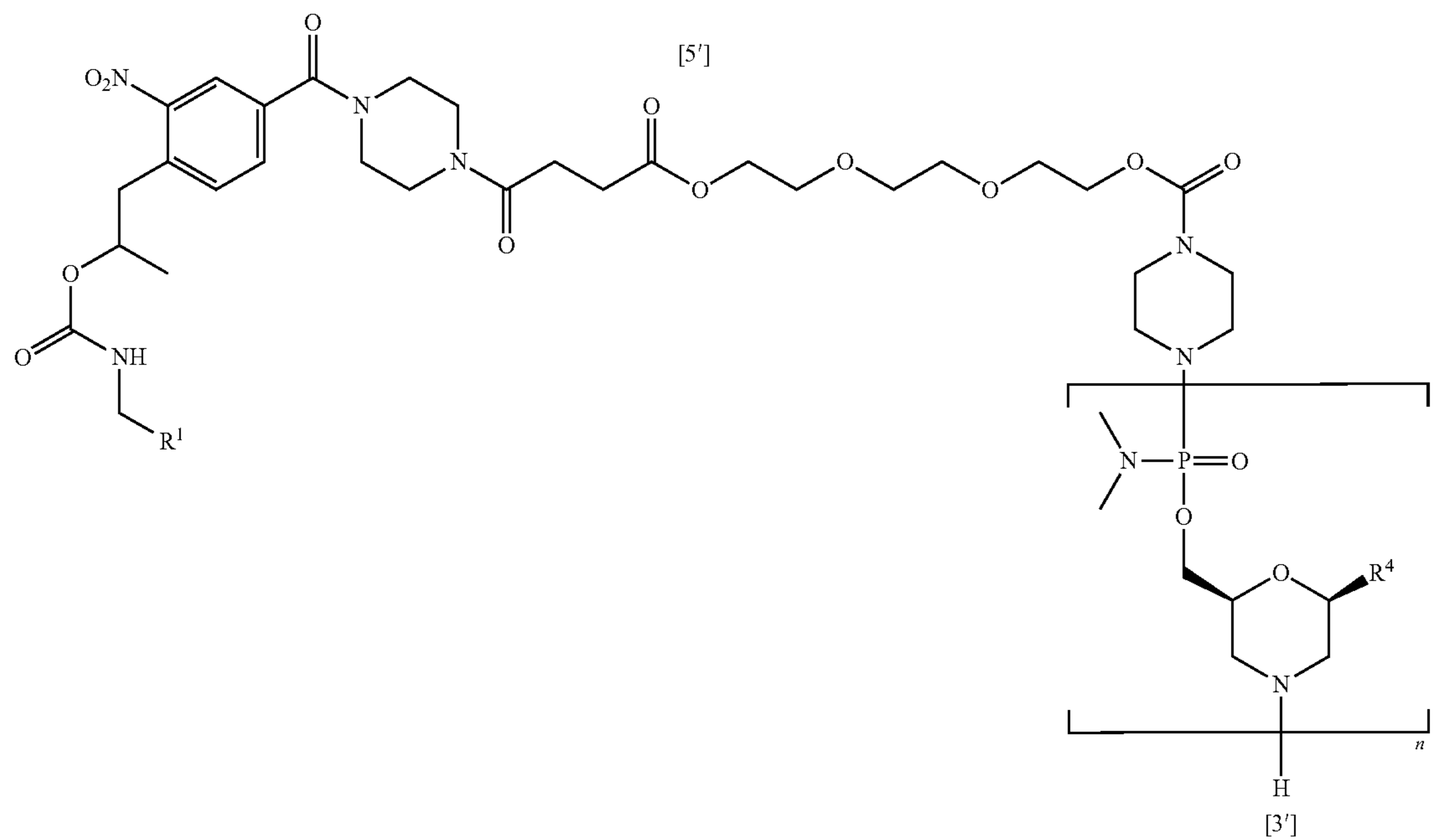
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65



173

In one embodiment, the compound of Formula (A10) is of Formula (A10a):



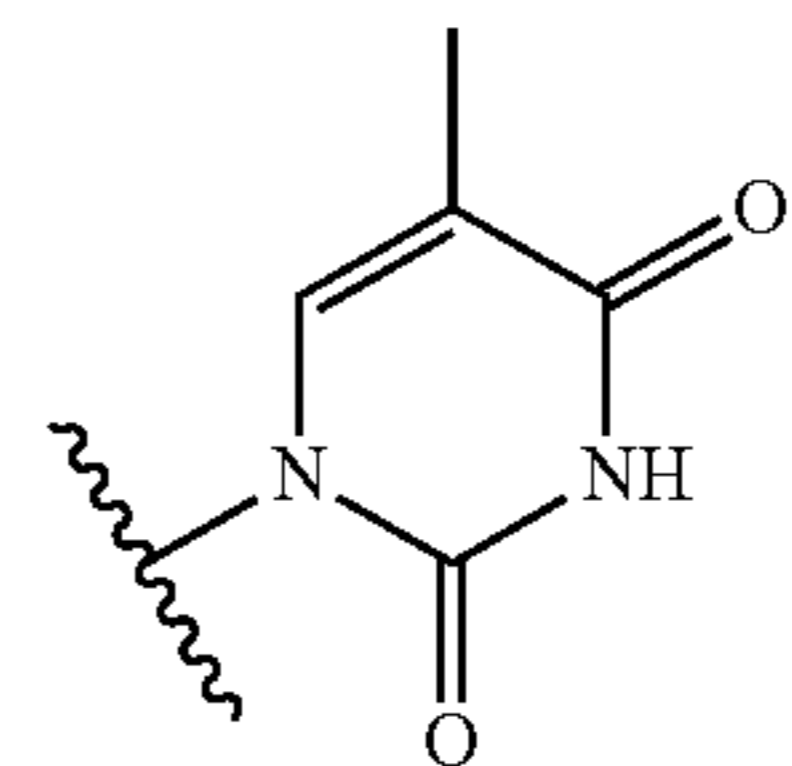
(A10a)

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or a pharmaceutically acceptable salt thereof, wherein:
 n is an integer from 10 to 40;
 R¹ is a support-medium; and
 R⁴ is, independently at each occurrence, selected from the group consisting of:

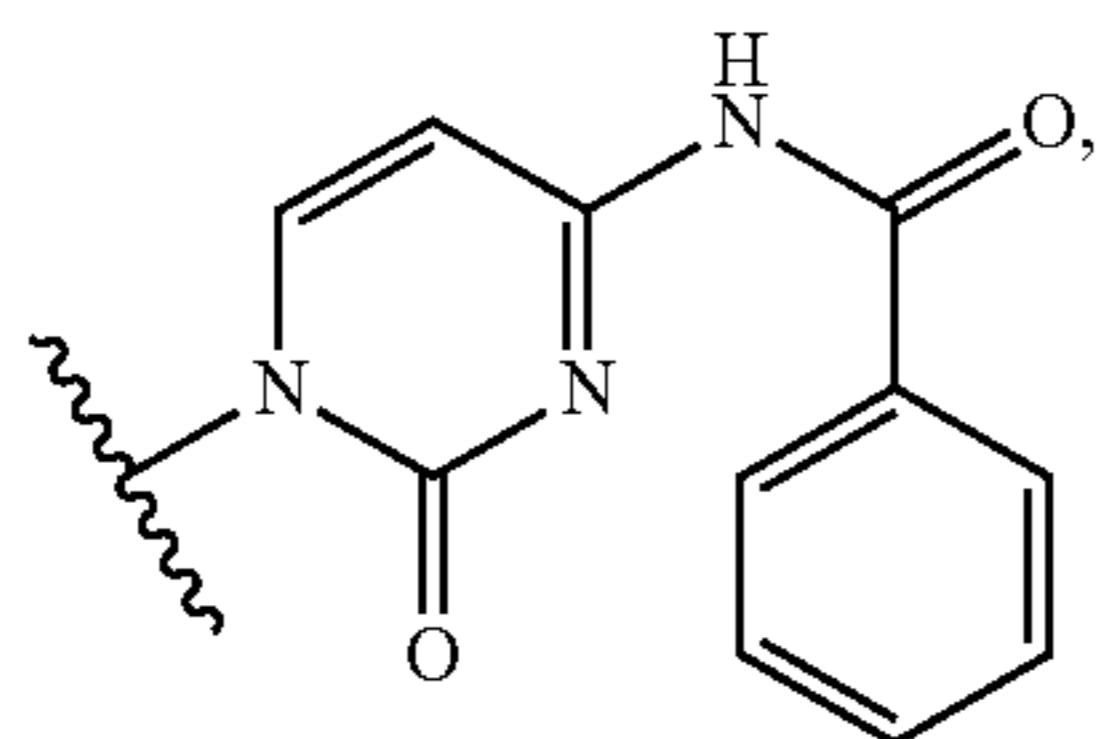
-continued

(T)



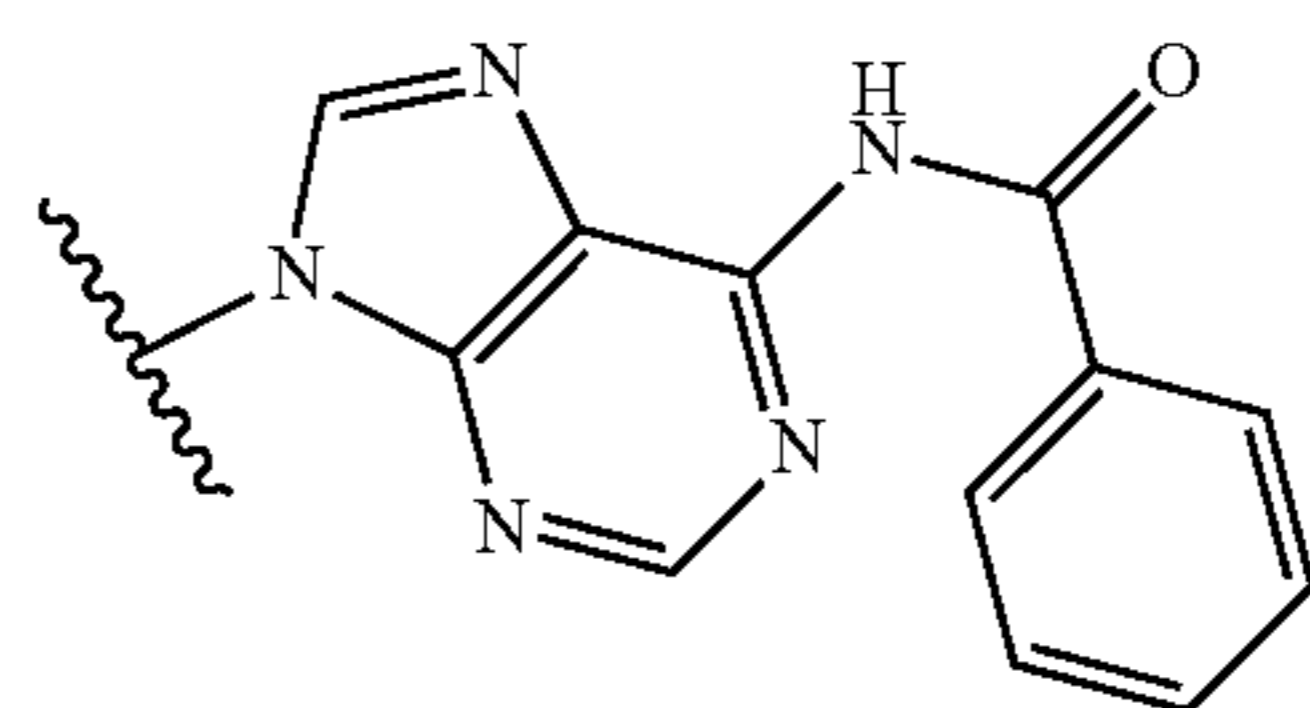
35

(PC) 40

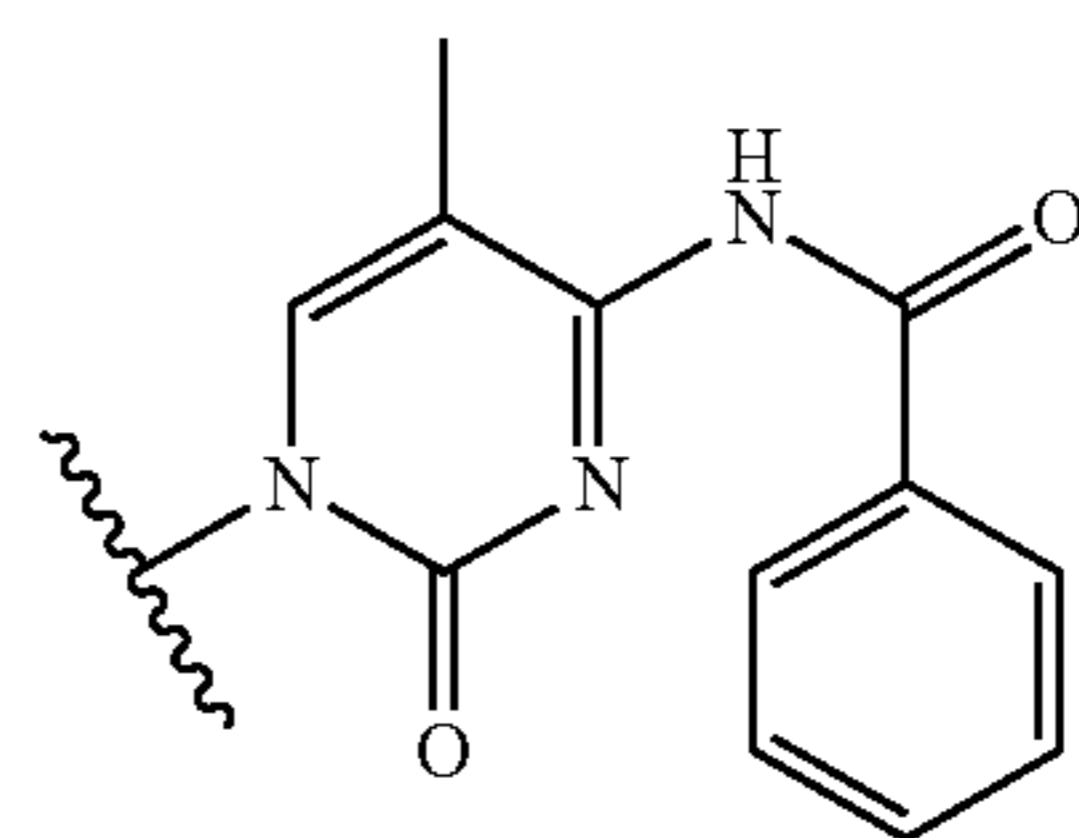


45

(PA)

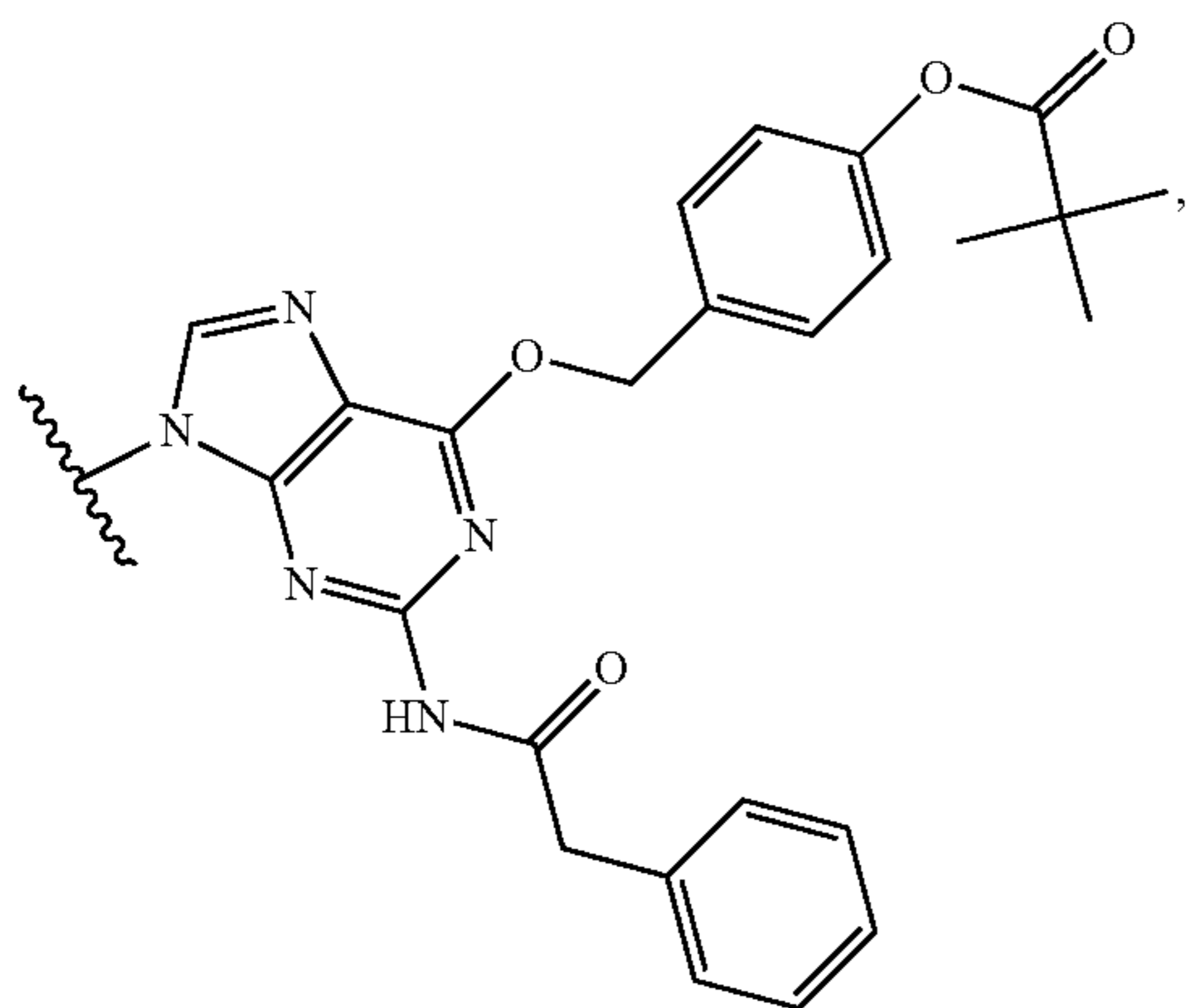


(DPG) 50



55

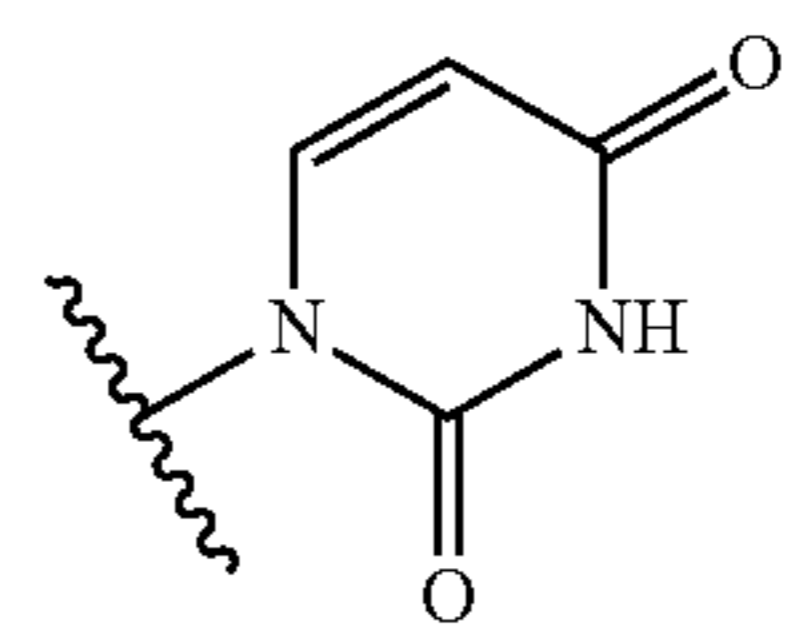
(P5mC)



60

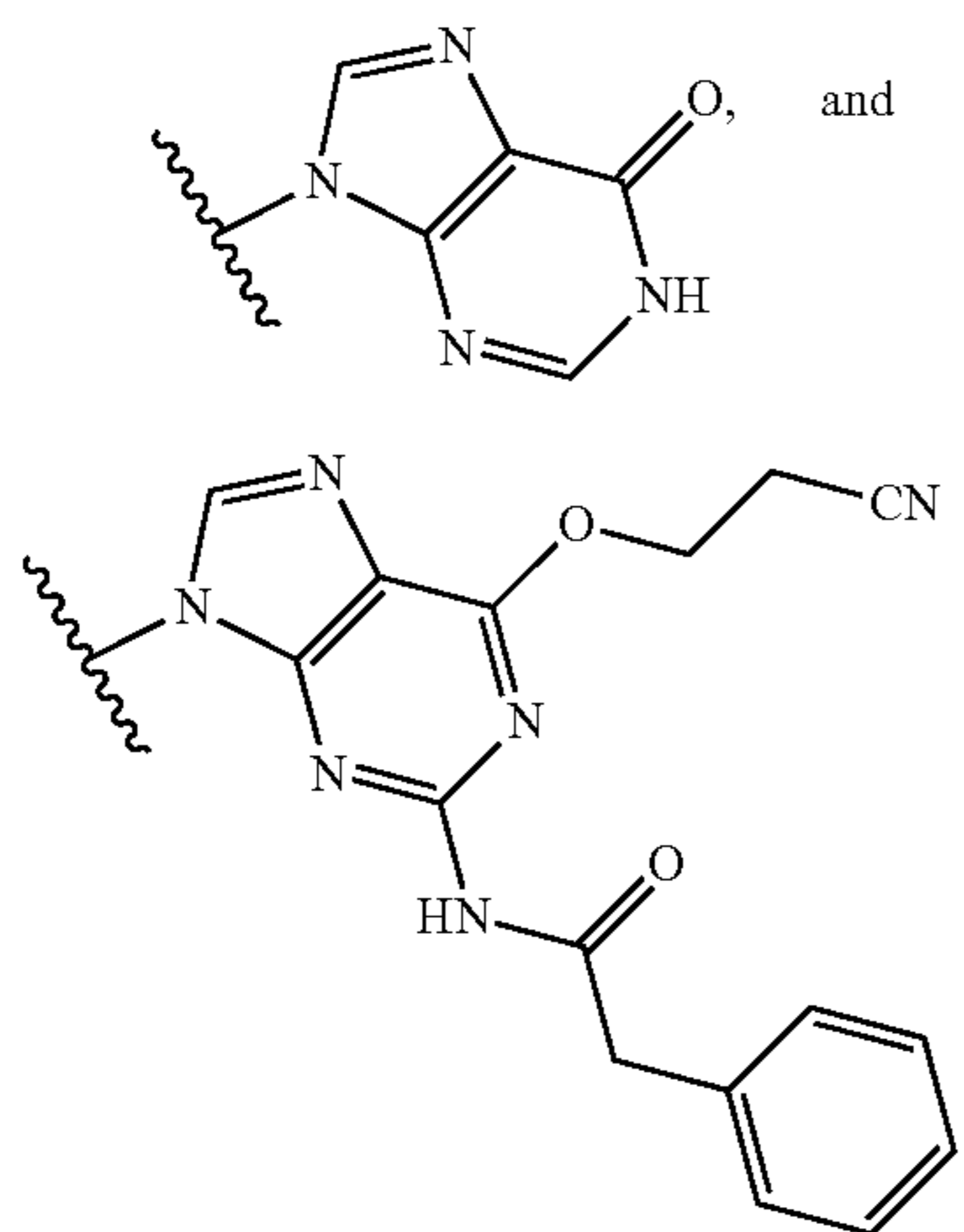
(U)

65



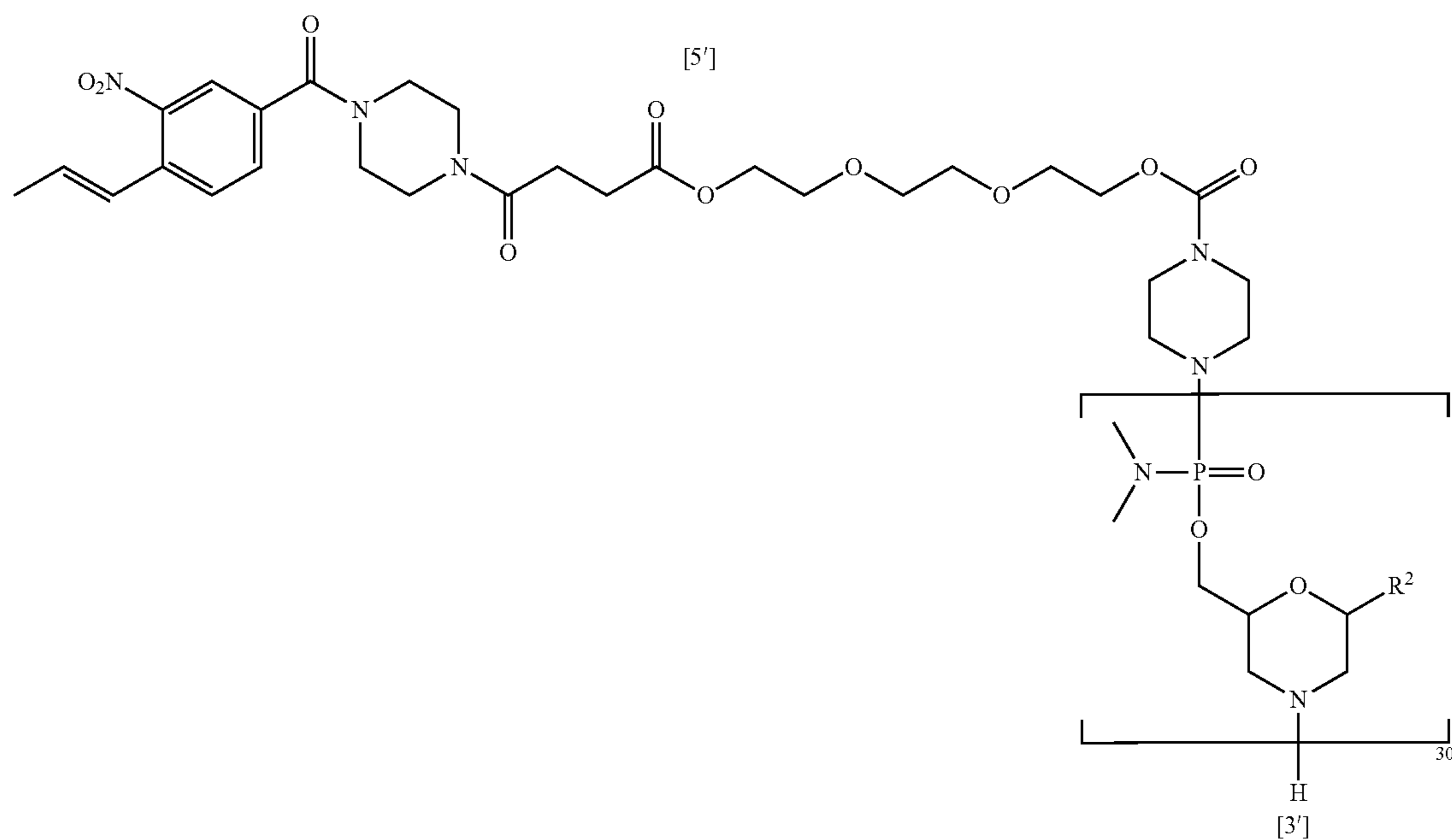
175

-continued

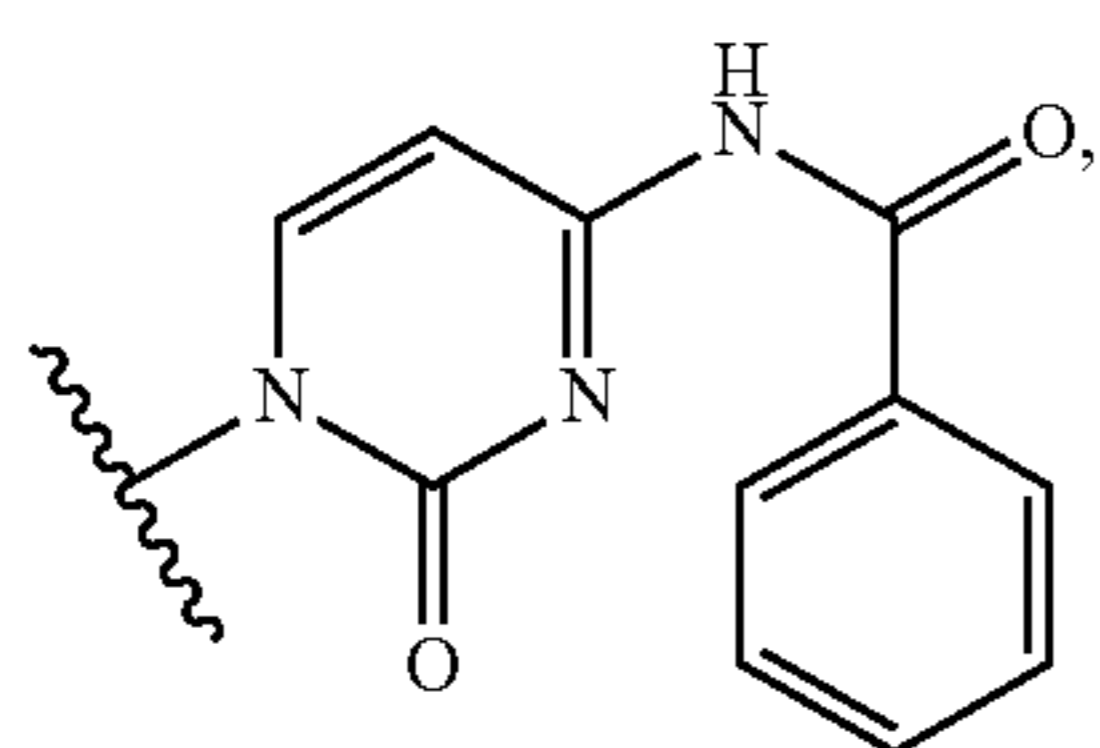


In another embodiment of these compounds, the support-medium comprises polystyrene with 1% crosslinked divinylbenzene.

In another aspect, provided herein is a compound of Formula (XI):

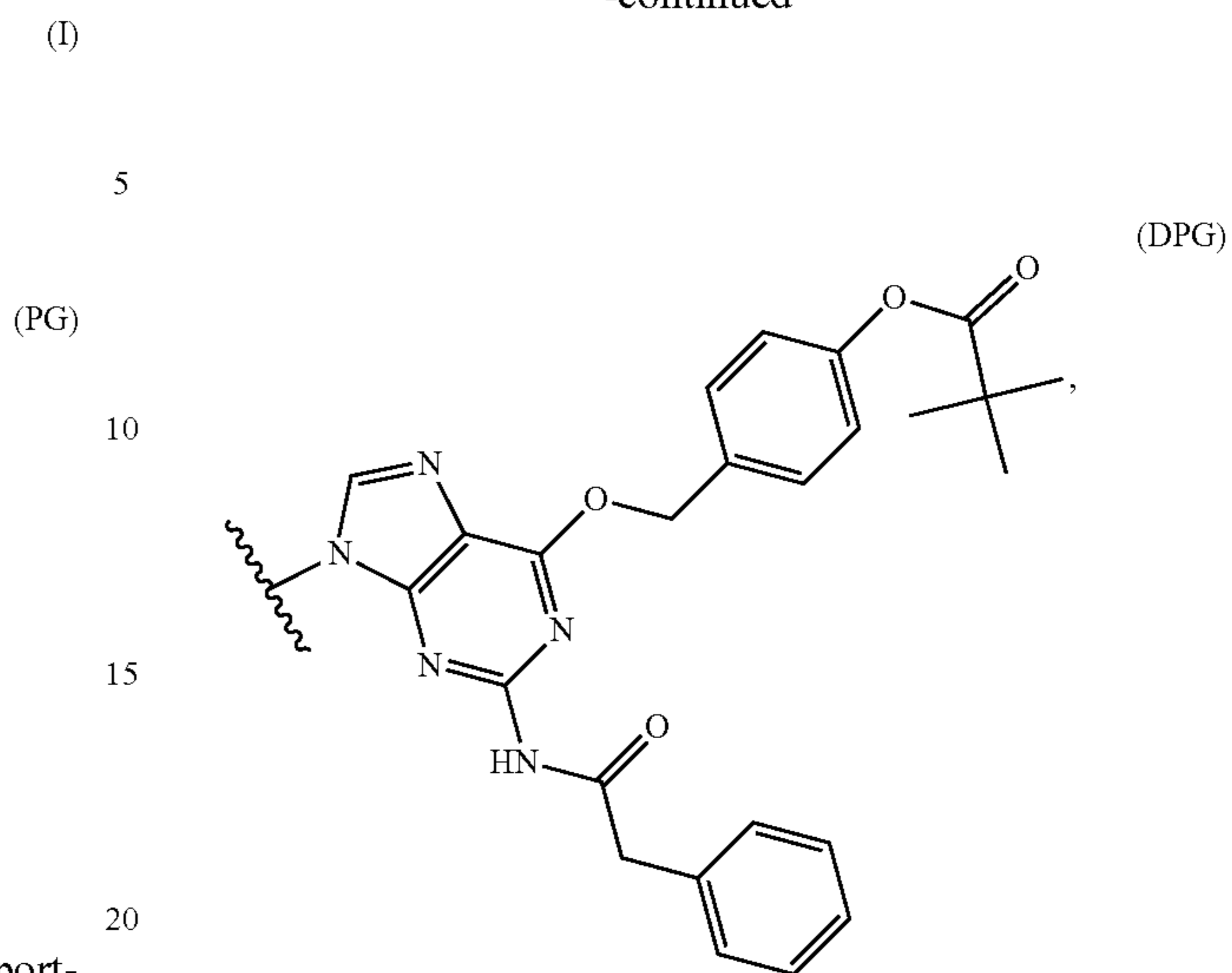


or a pharmaceutically acceptable salt thereof, wherein:
 R^2 is, independently at each occurrence, selected from the group consisting of:

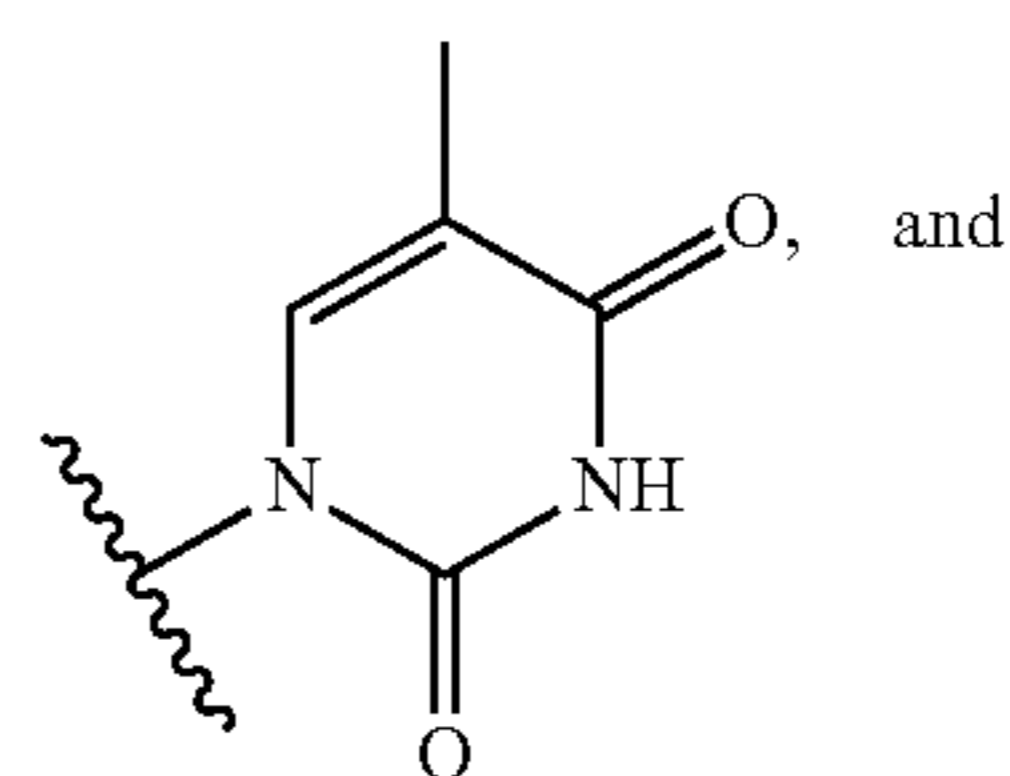


176

-continued

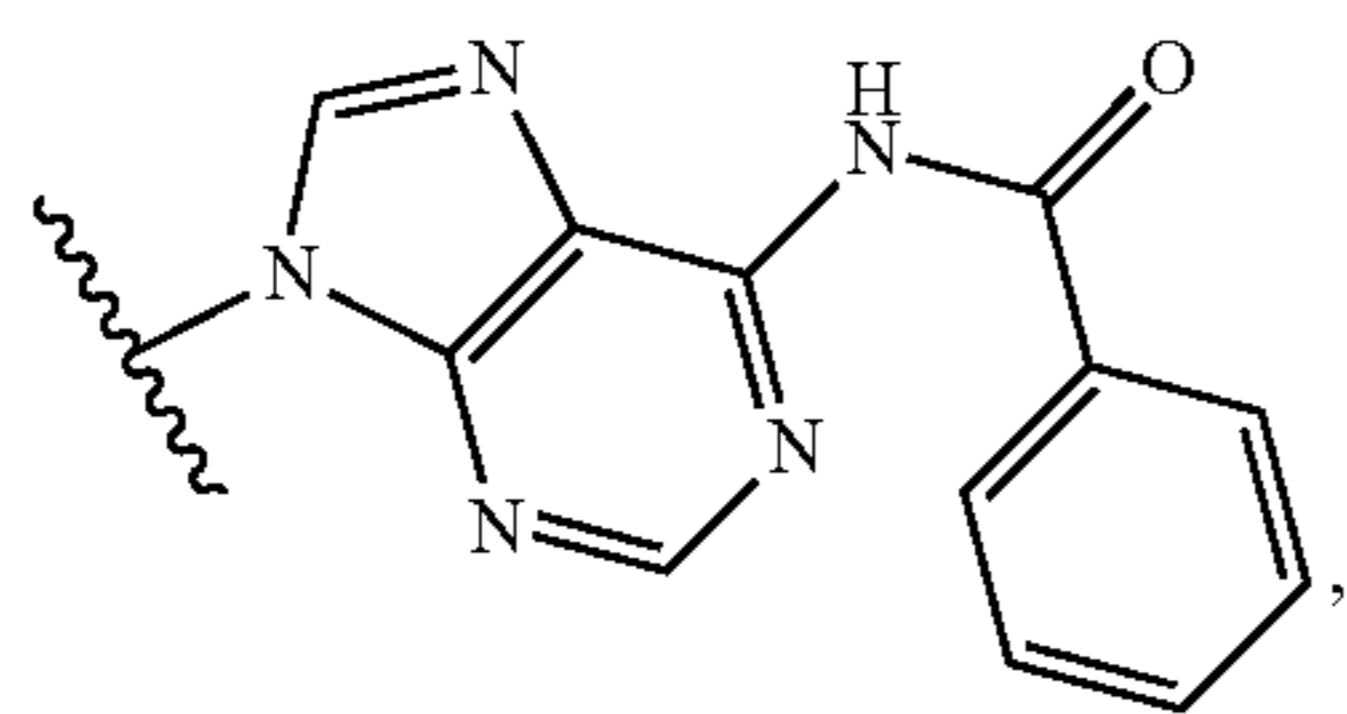


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177

-continued



and

wherein R² is at each position from 1 to 30 and 5' to 3':

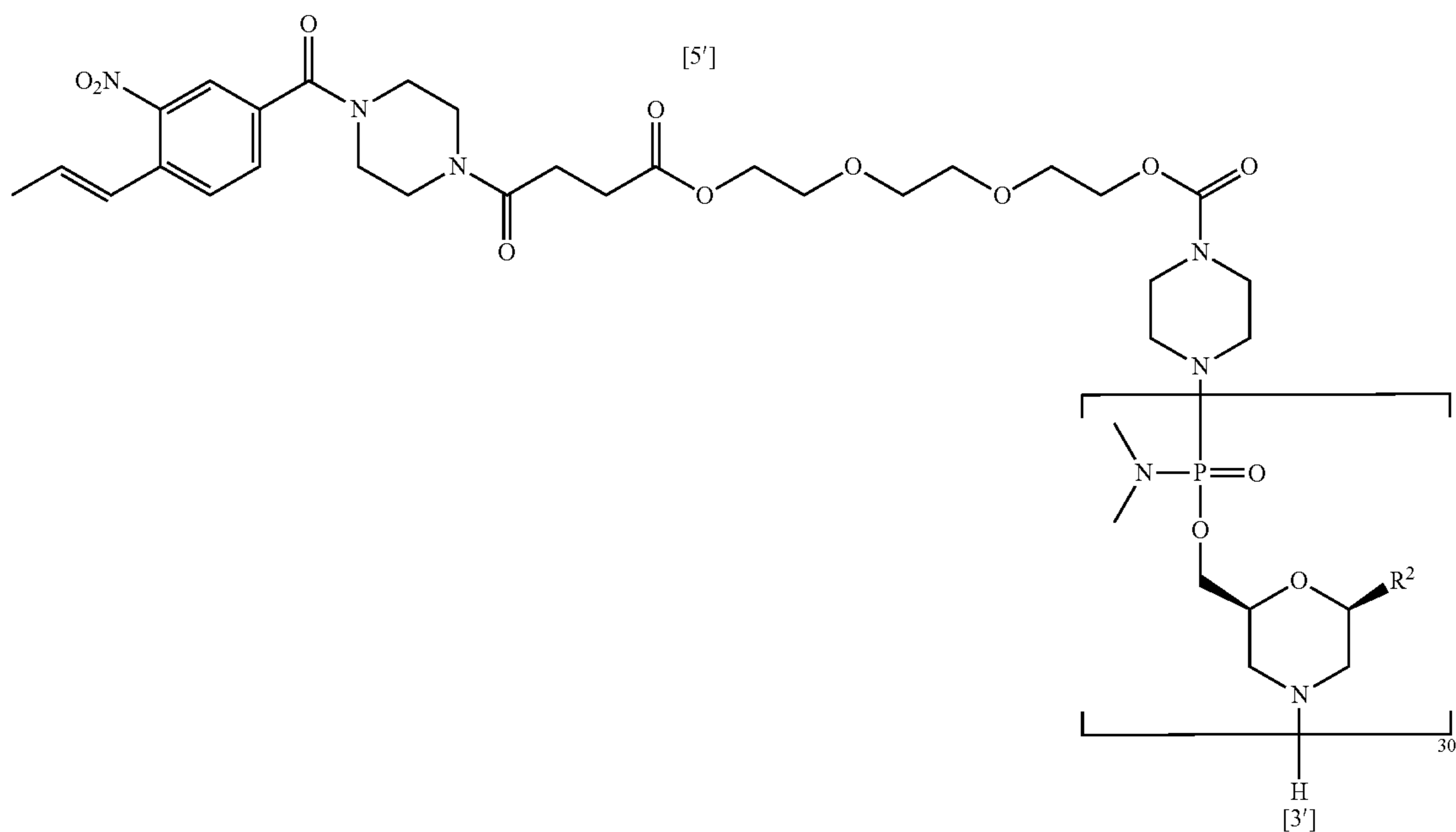
Position No. 5' to 3'	R ²
1	PC
2	T
3	PC
4	PC
5	PA
6	PA
7	PC

178

-continued

(PA)	Position No. 5' to 3'	R ²	
5	19	T	
	20	DPG	
	21	DPG	
	22	PC	
	10	23	PA
		24	T
		25	T
15	26	T	
	27	PC	
	28	T	
20	29	PA	
	30	DPG	

In one embodiment, the compound of Formula (XI) is of Formula (XIa):



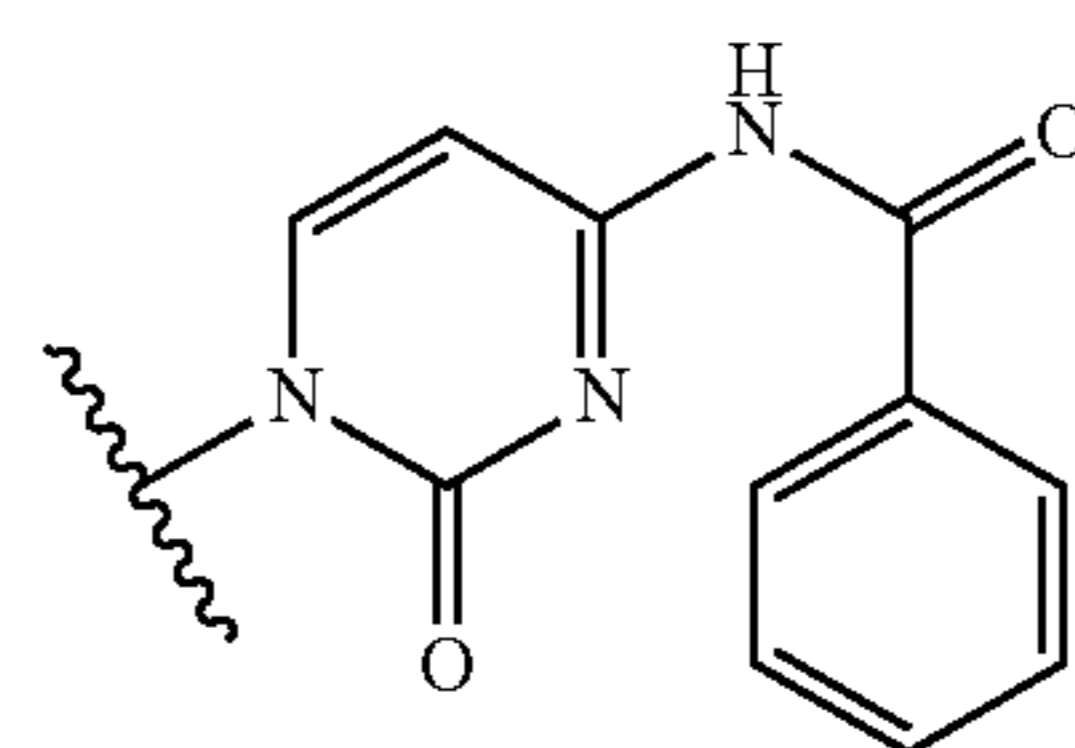
(XIa)

-continued

Position No. 5' to 3'	R ²
8	PA
9	T
10	PC
11	PA
12	PA
13	DPG
14	DPG
15	PA
16	PA
17	DPG
18	PA

or a pharmaceutically acceptable salt thereof, wherein:

R² is, independently at each occurrence, selected from the group consisting of:



(PC)

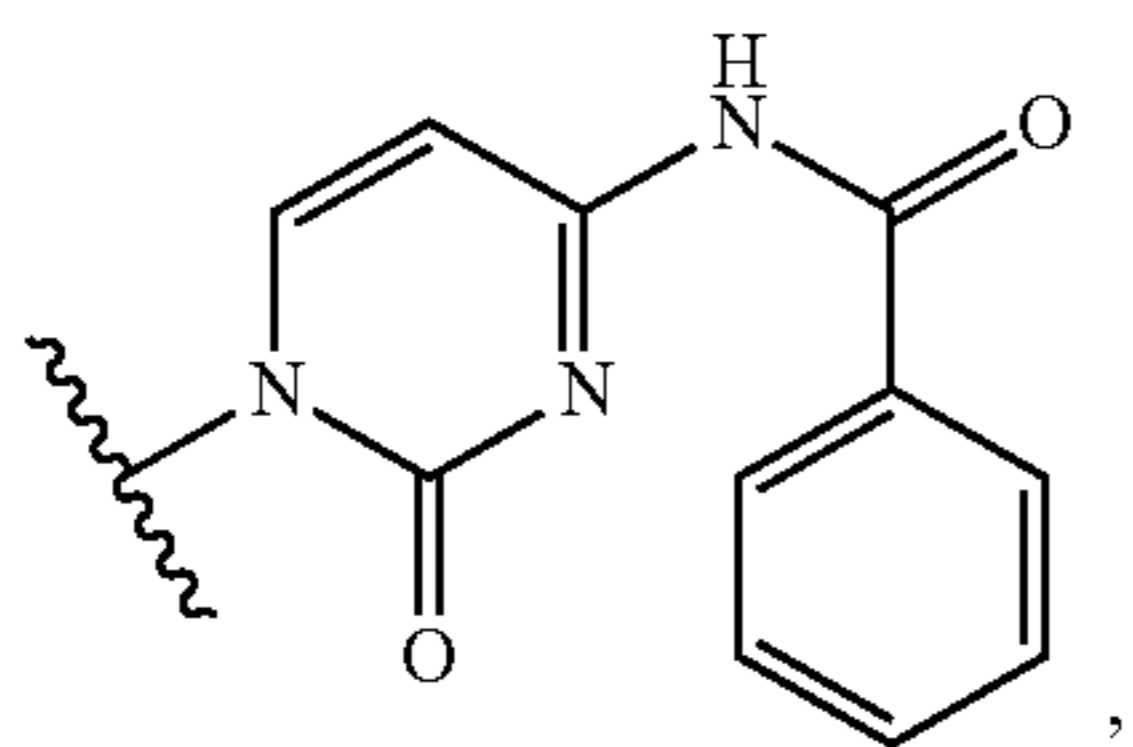
181

or a pharmaceutically acceptable salt thereof, wherein:

n is an integer from 10 to 40;

R¹ is a support-medium; and

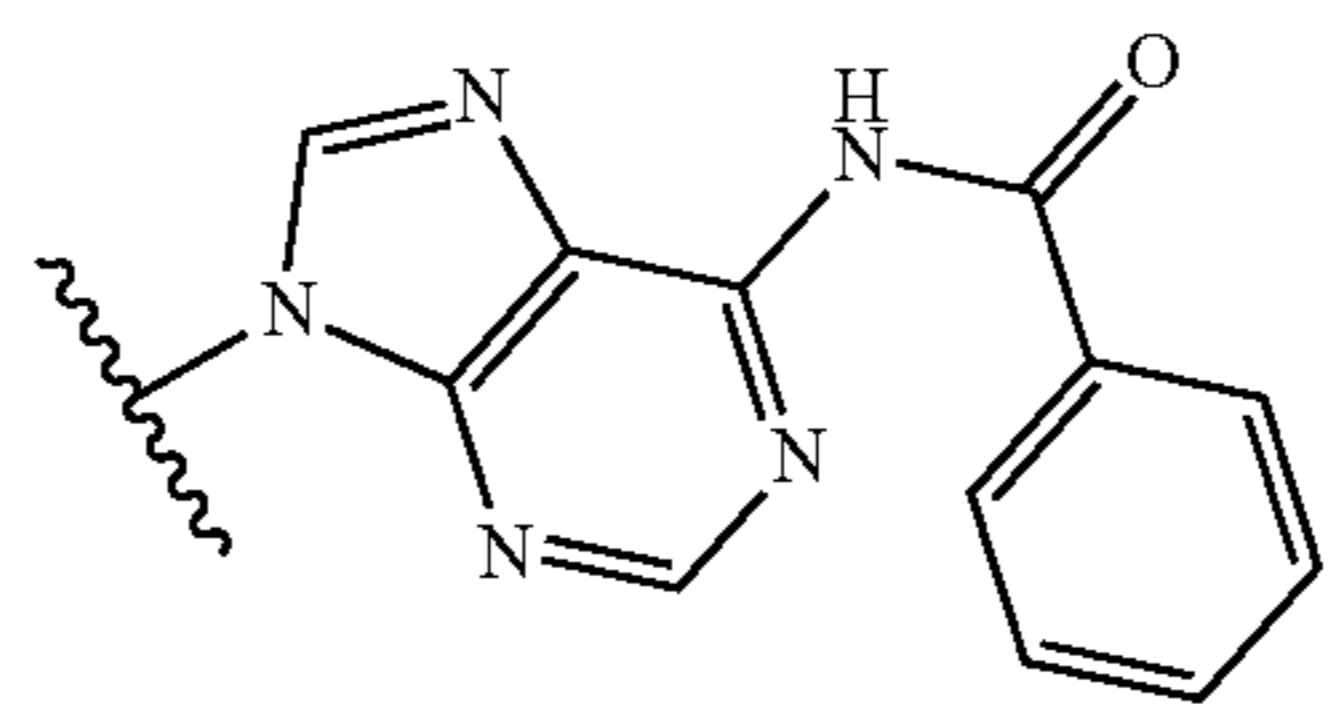
R⁴ is, independently at each occurrence, selected from the group consisting of:



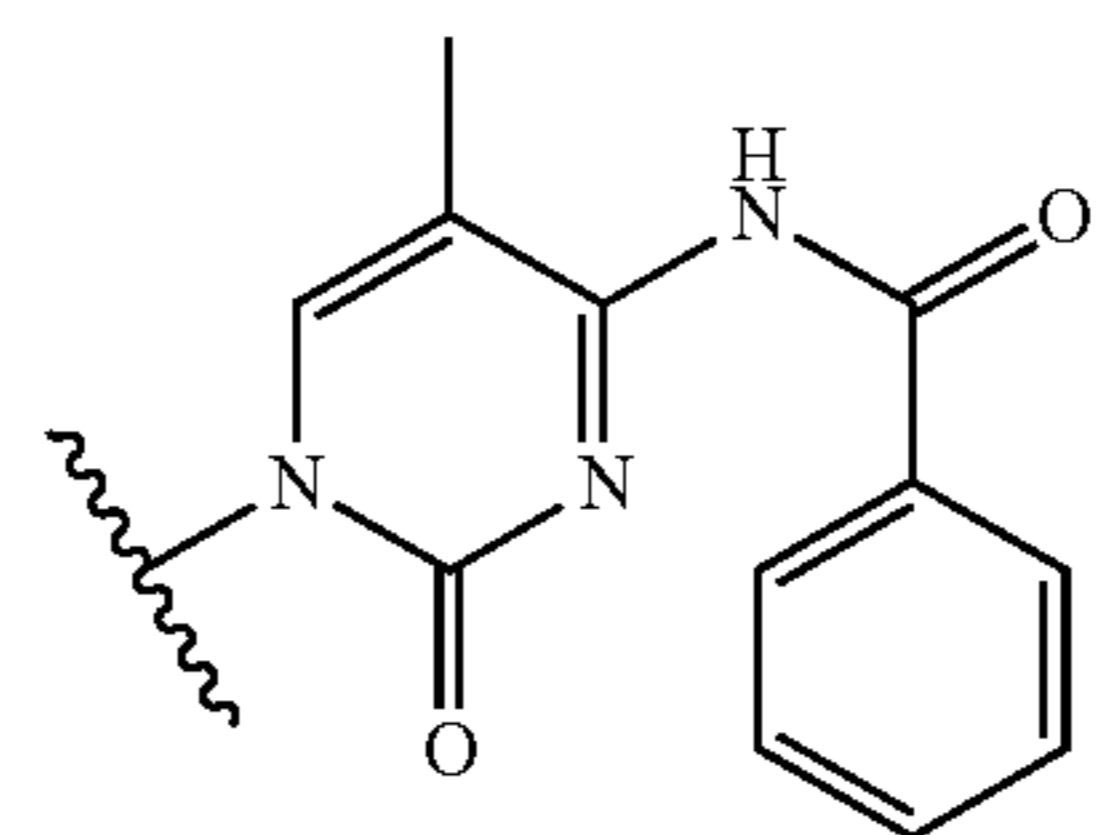
(PC)

10

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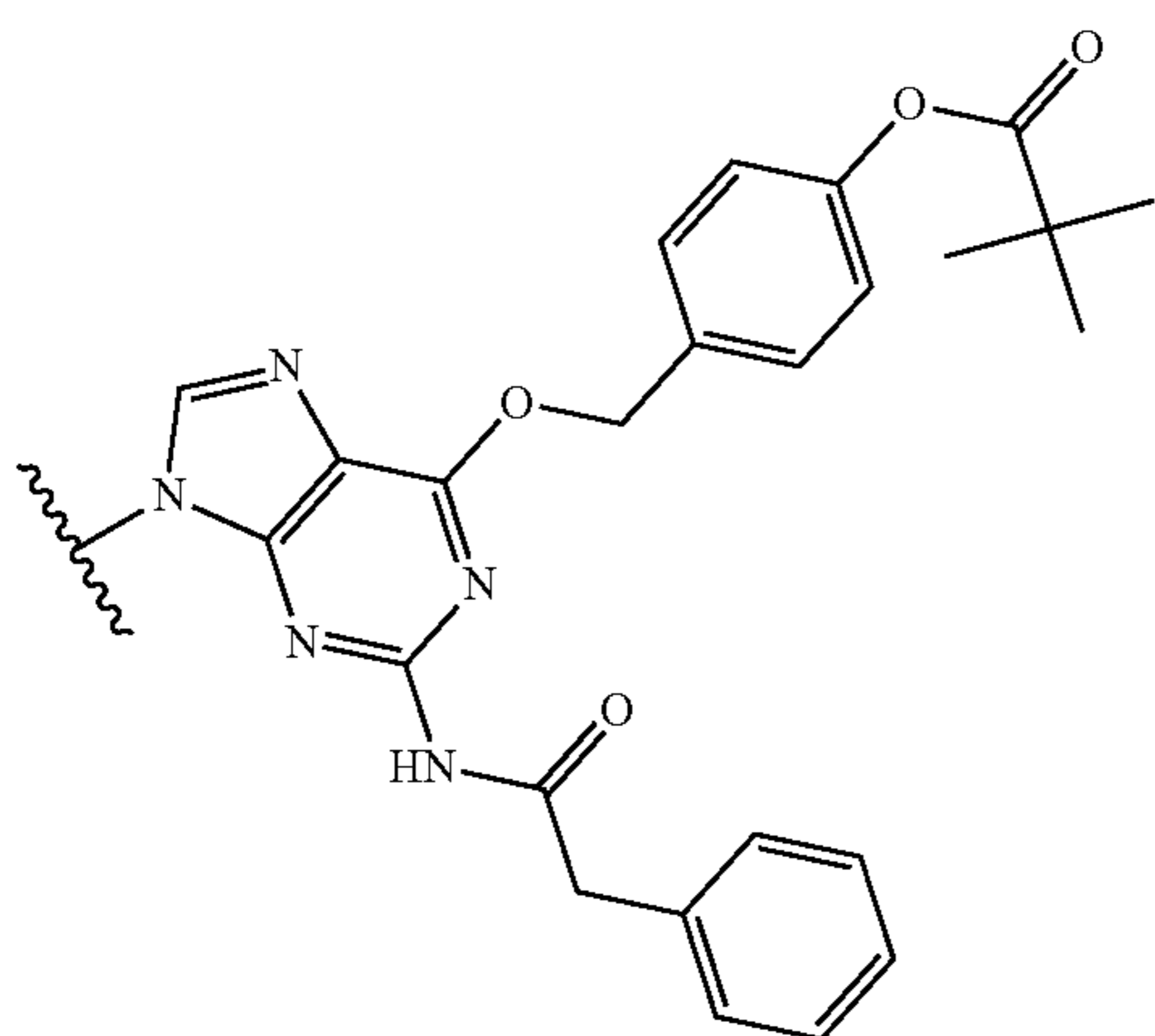
(PA)



(P5mC)

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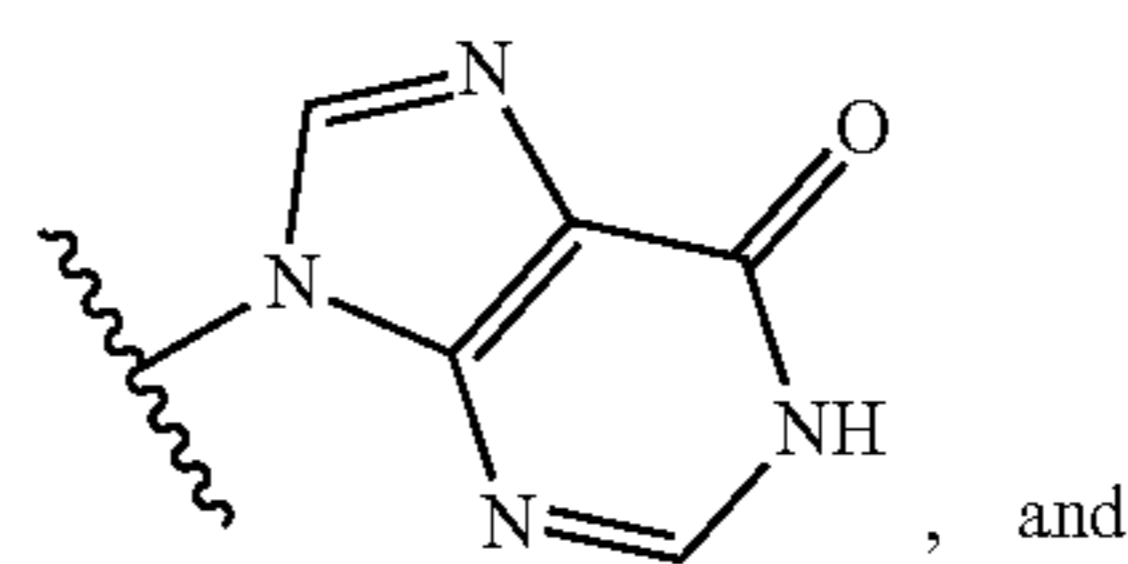
(DPG)

30

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40

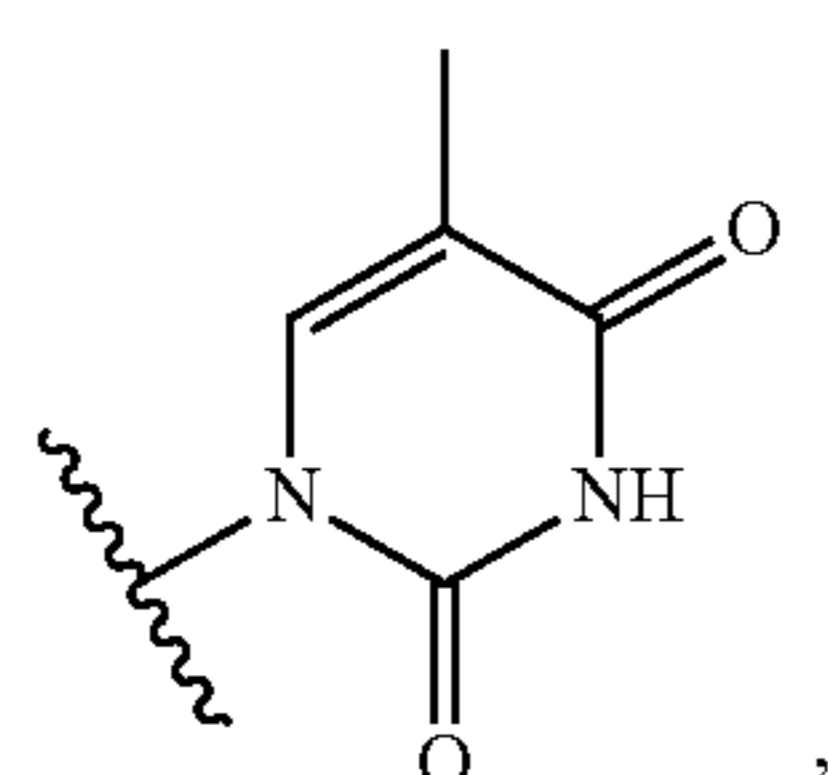
45



(U)

(I)

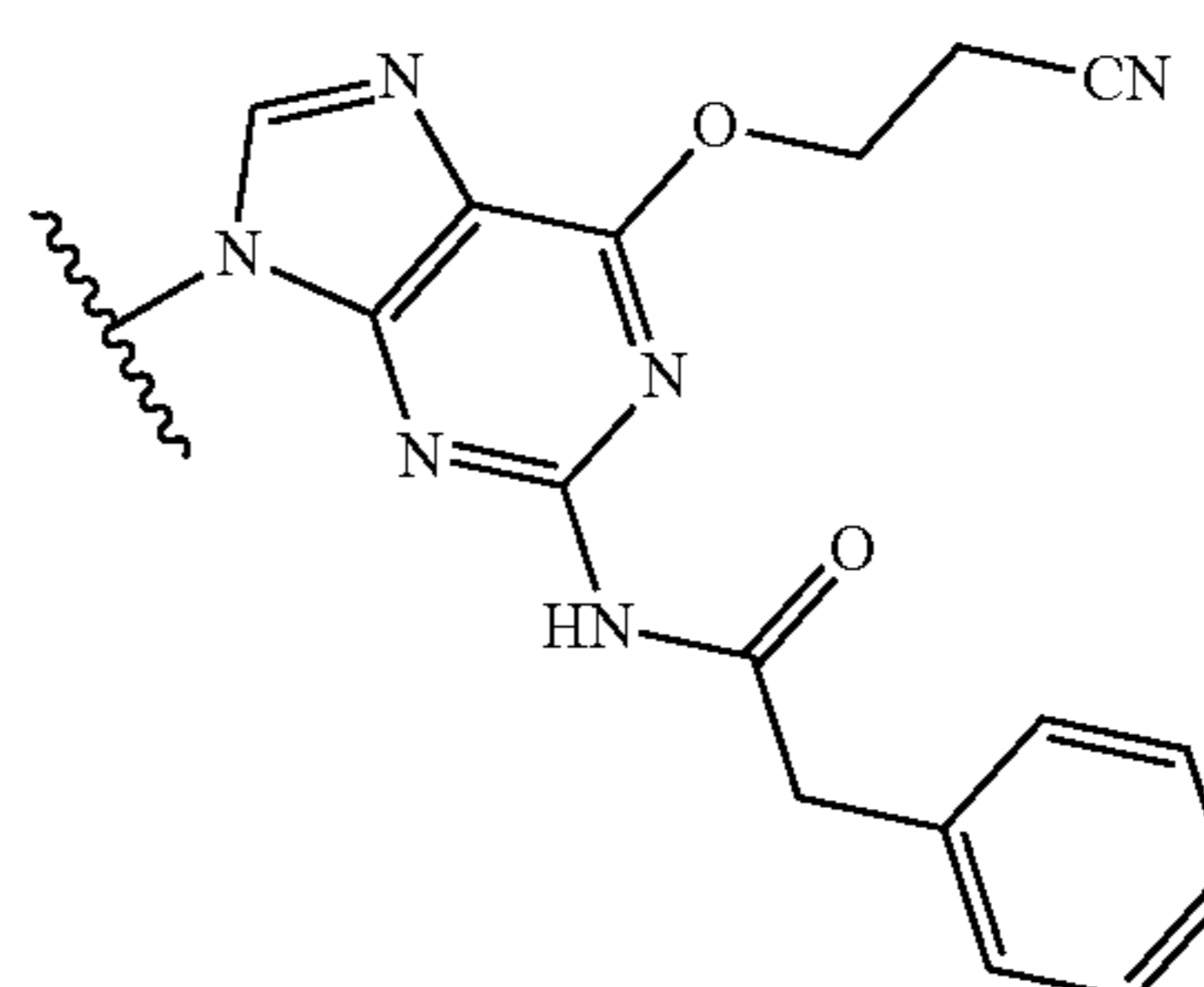
50



(T)

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65

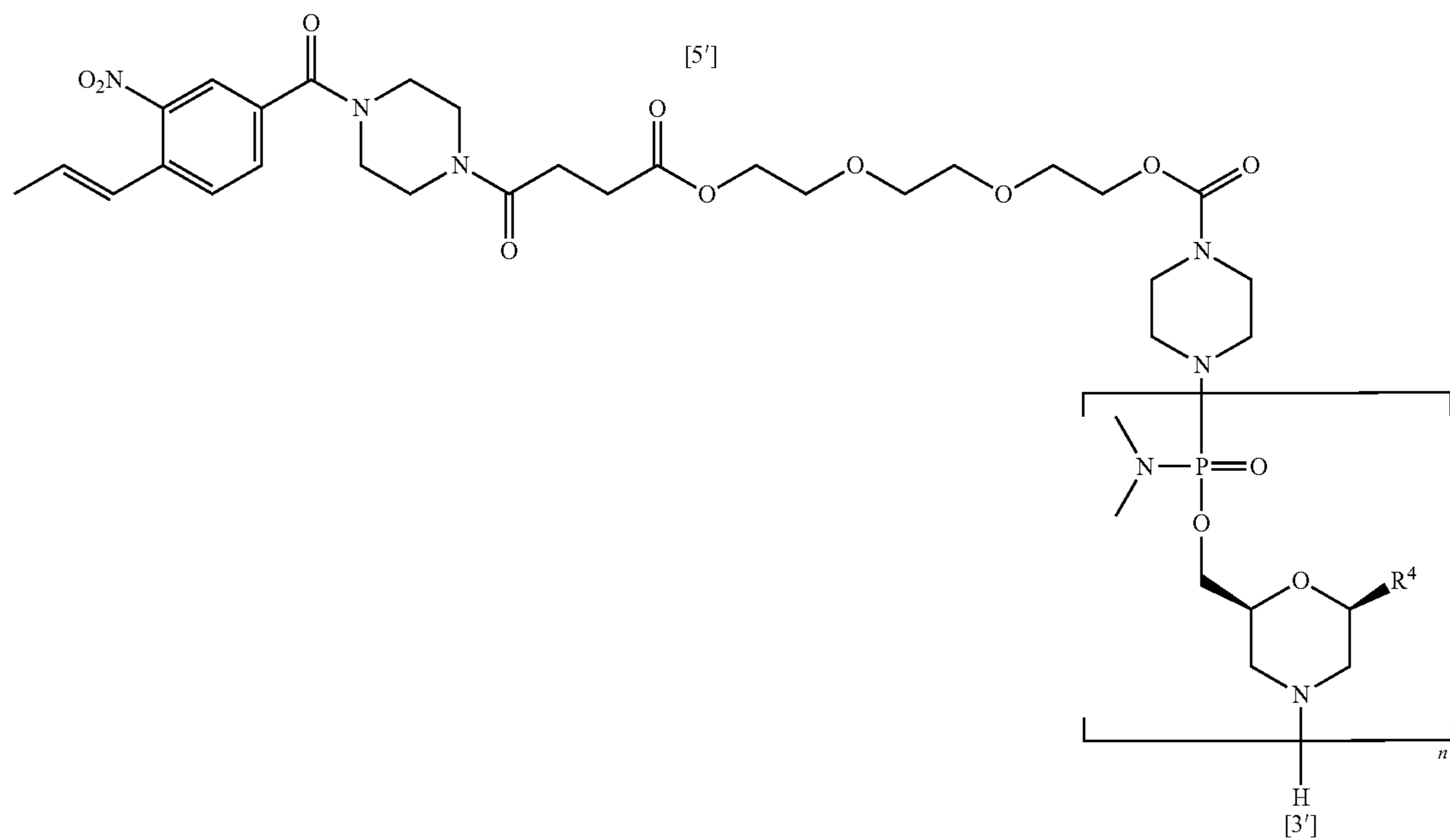


(PG)

55

183

In one embodiment, the compound of Formula (A11) is of formula (A11a):

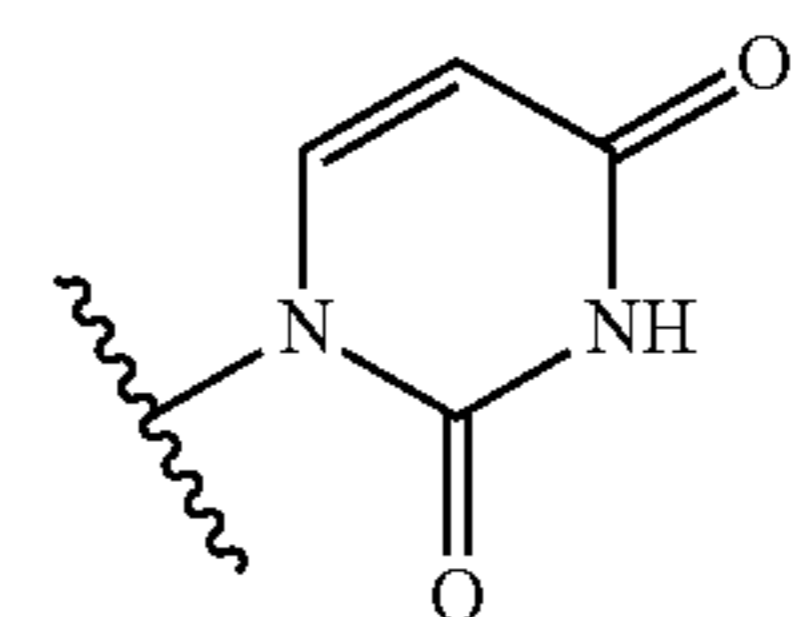
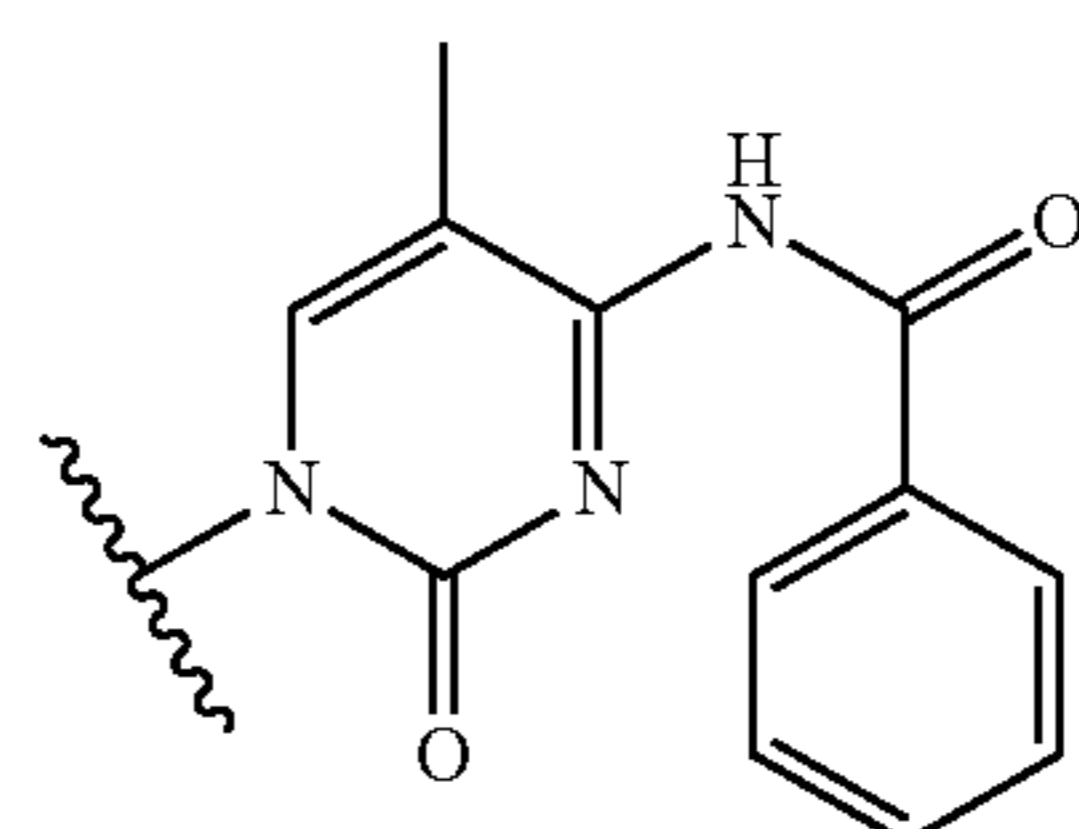
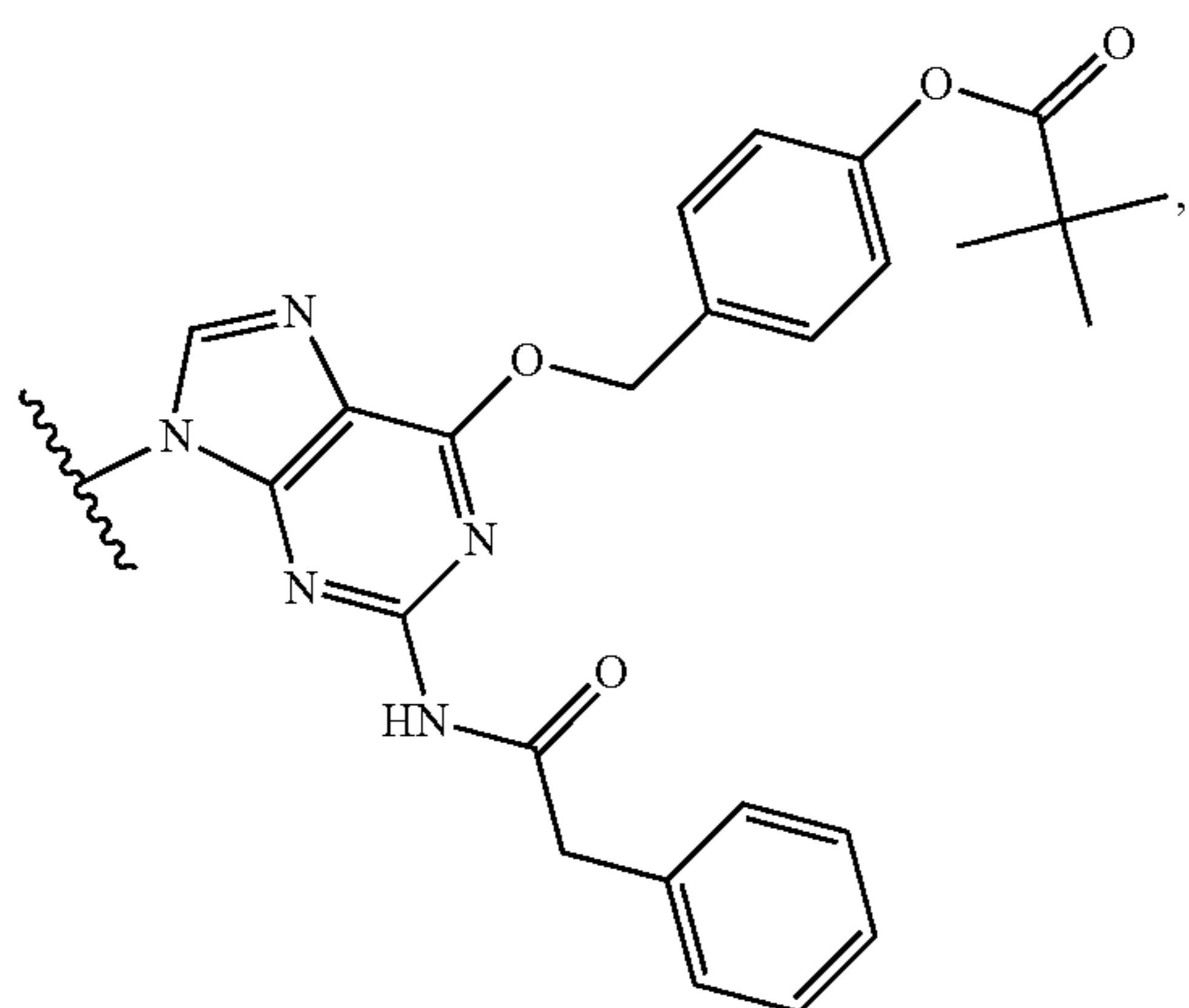
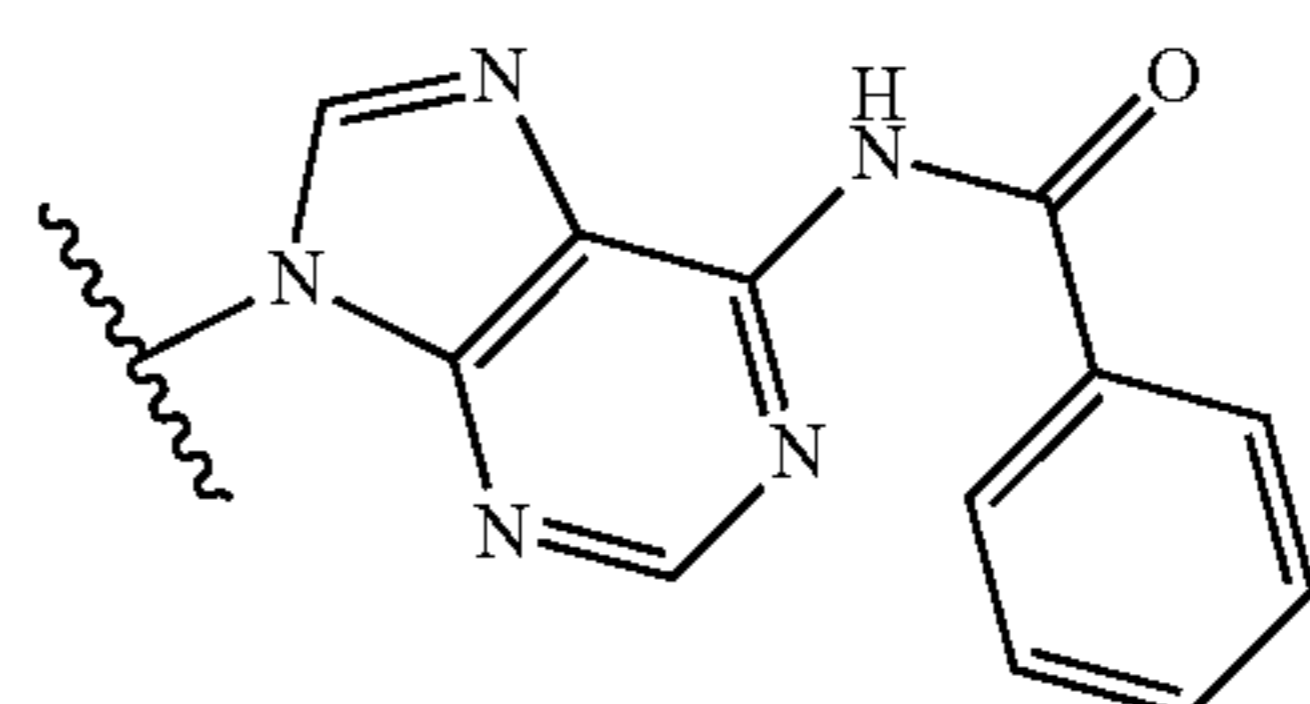
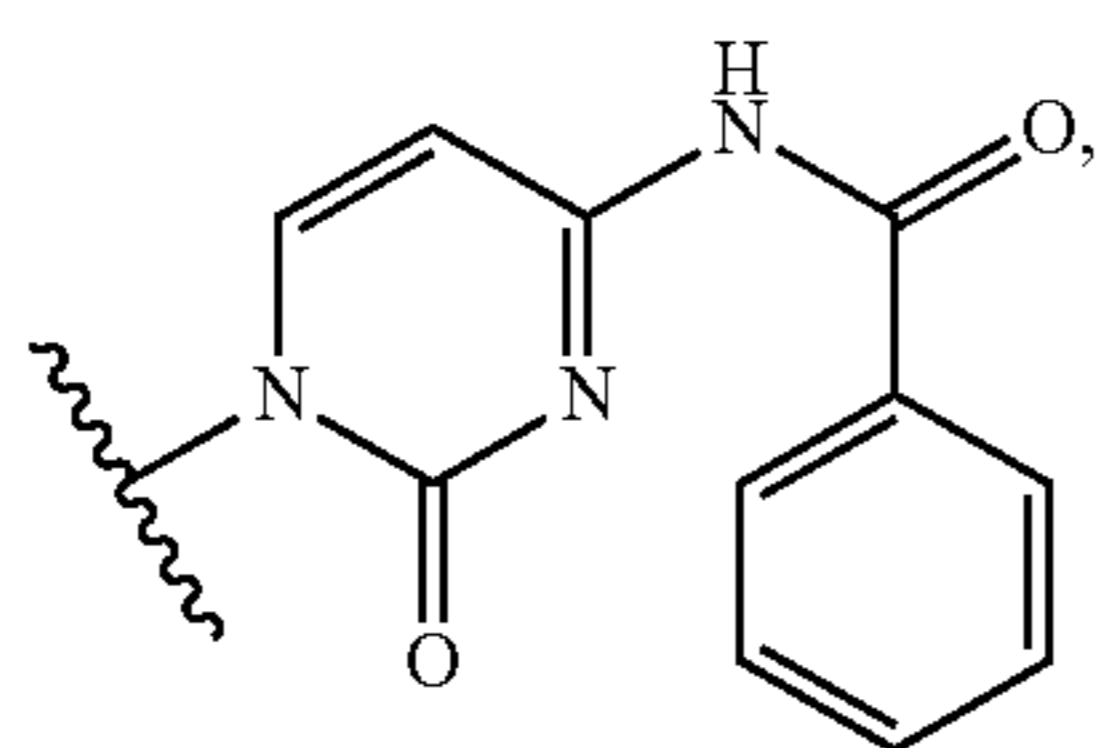


or a pharmaceutically acceptable salt thereof, wherein:

n is an integer from 10 to 40;

R^1 is a support-medium; and

R^4 is, independently at each occurrence, selected from the group consisting of:



-continued

(T)

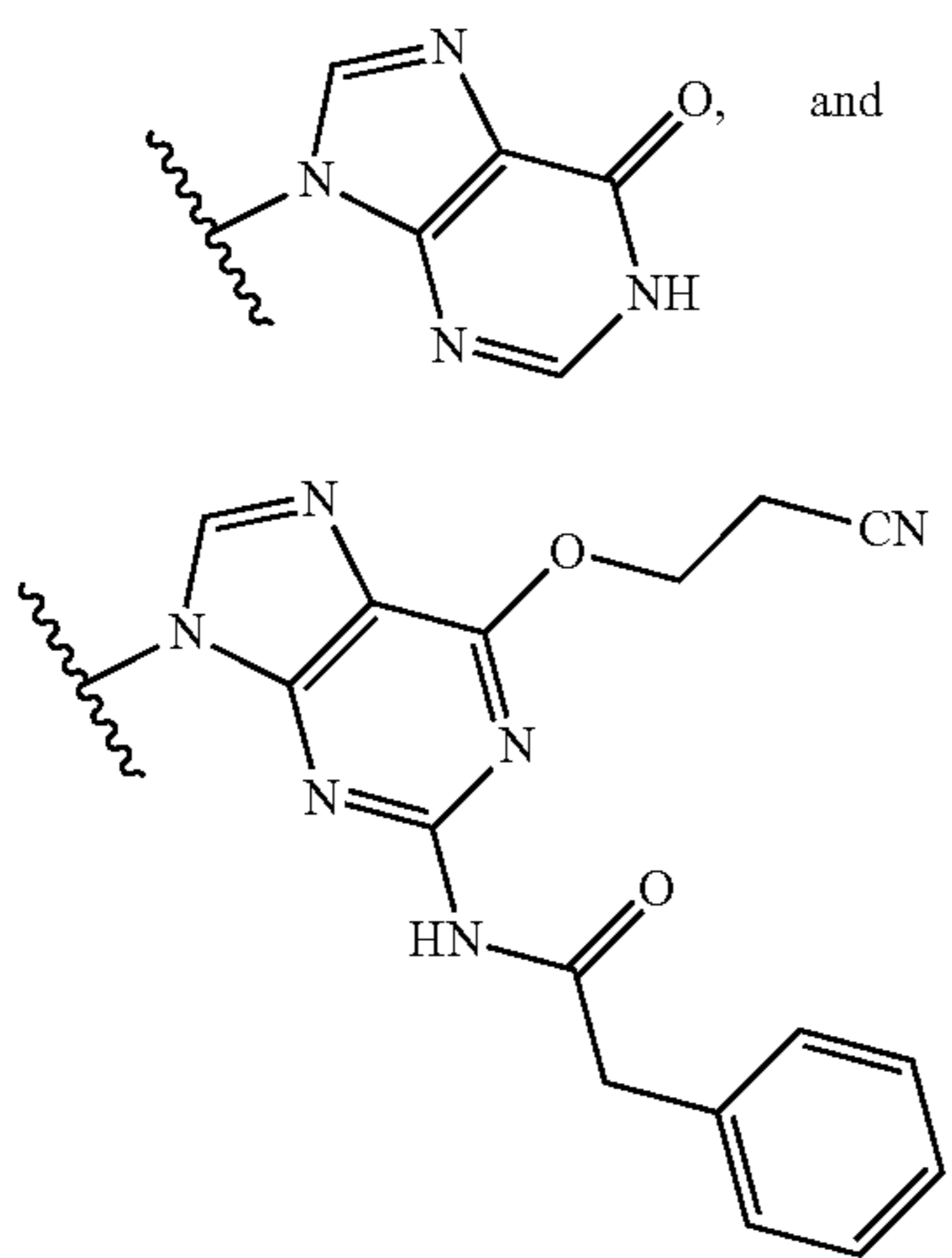
(PA)

(P5mC)

(U)

185

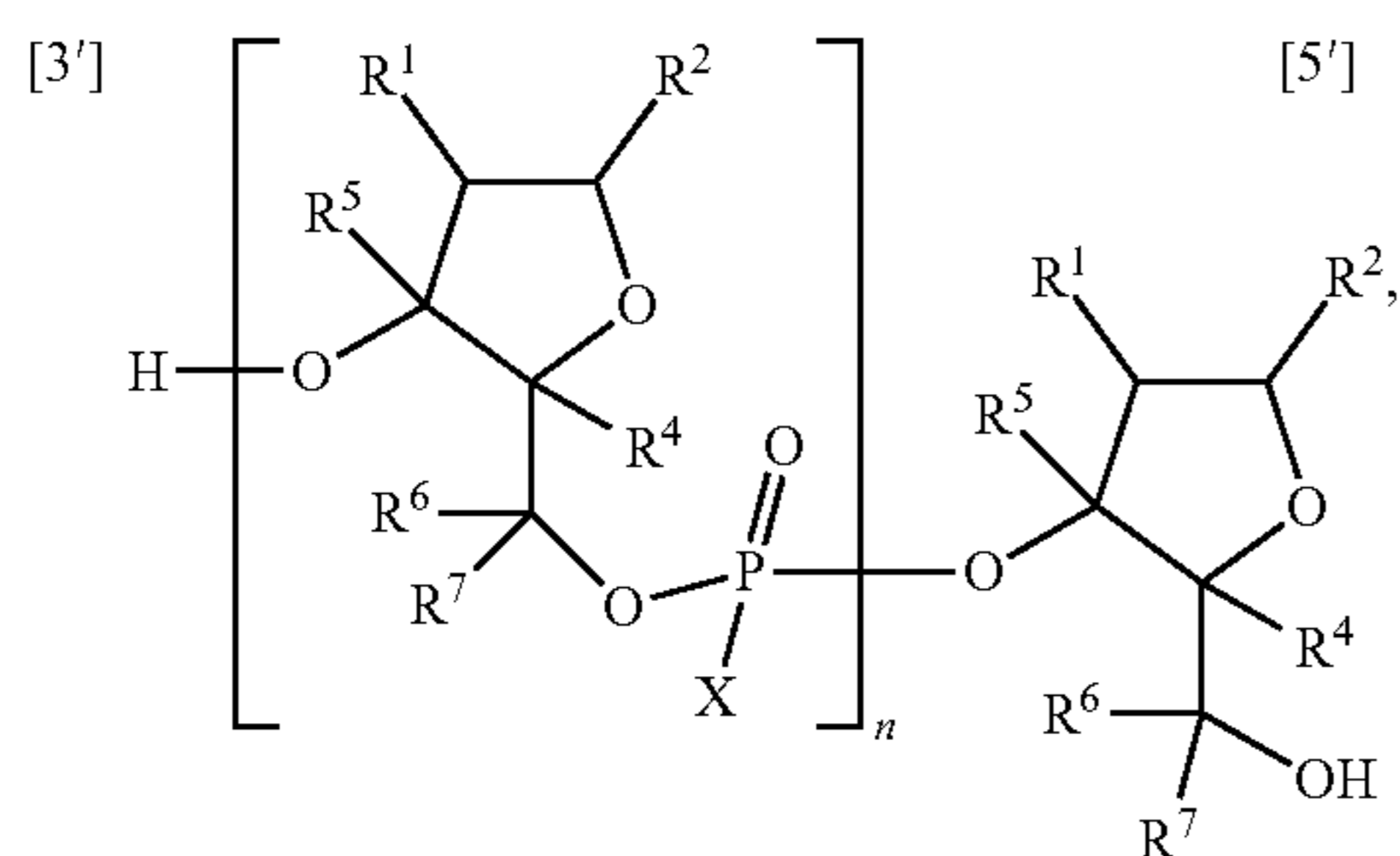
-continued



Oligomers

Oligomers prepared by the process of the disclosure may contain a variety of nucleotide analog subunits.

In another aspect of the disclosure is provided a process for preparing an oligomeric compound of Formula (D):



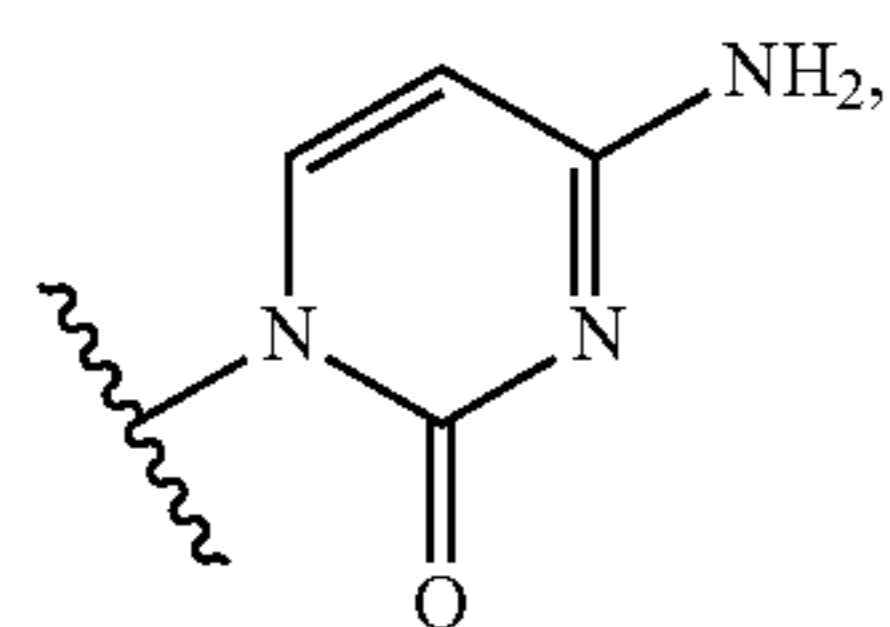
or a pharmaceutically acceptable salt thereof, wherein:

R¹ is selected from —O(C₁-C₆alkyl), halo, —O(C₁-C₆alkyl)-O(C₁-C₆alkyl), and —O(C₁-C₆alkyl)-C(O)—NH—C₁-C₆alkyl;

R² is H or R¹ and R² join to form a cycloalkyl or heterocyclic ring having from 4 to 7 ring atoms optionally containing an oxygen;

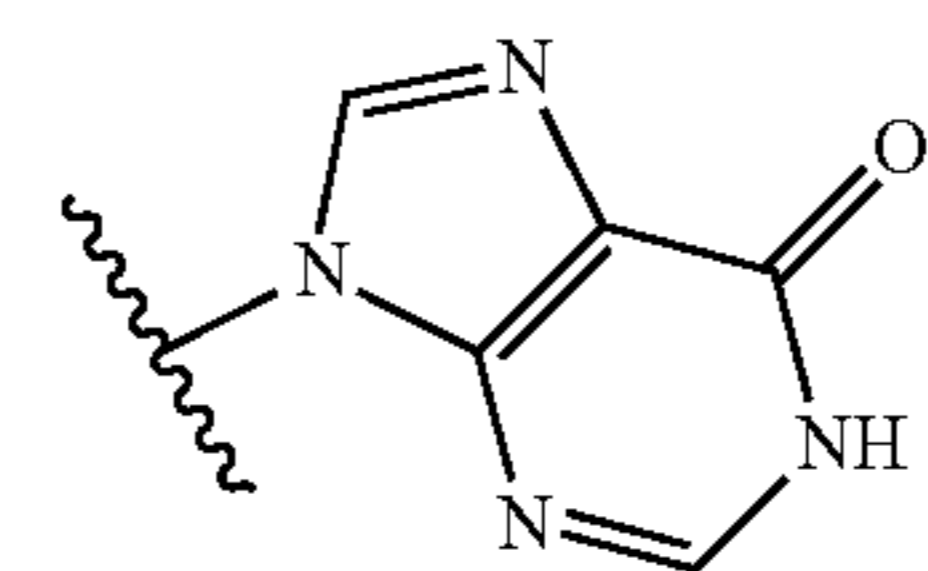
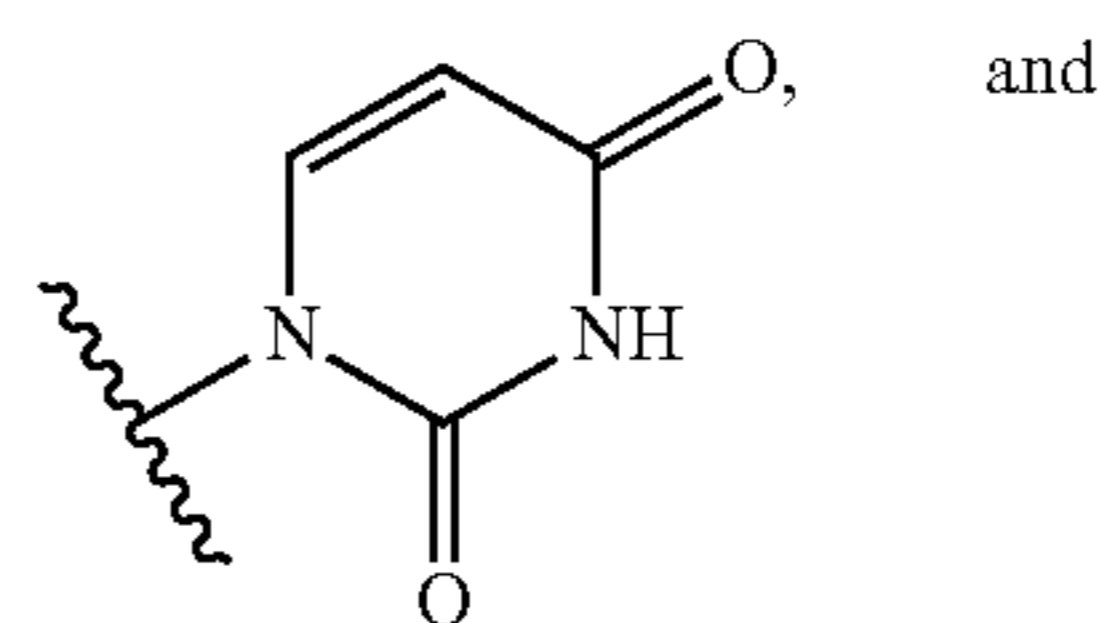
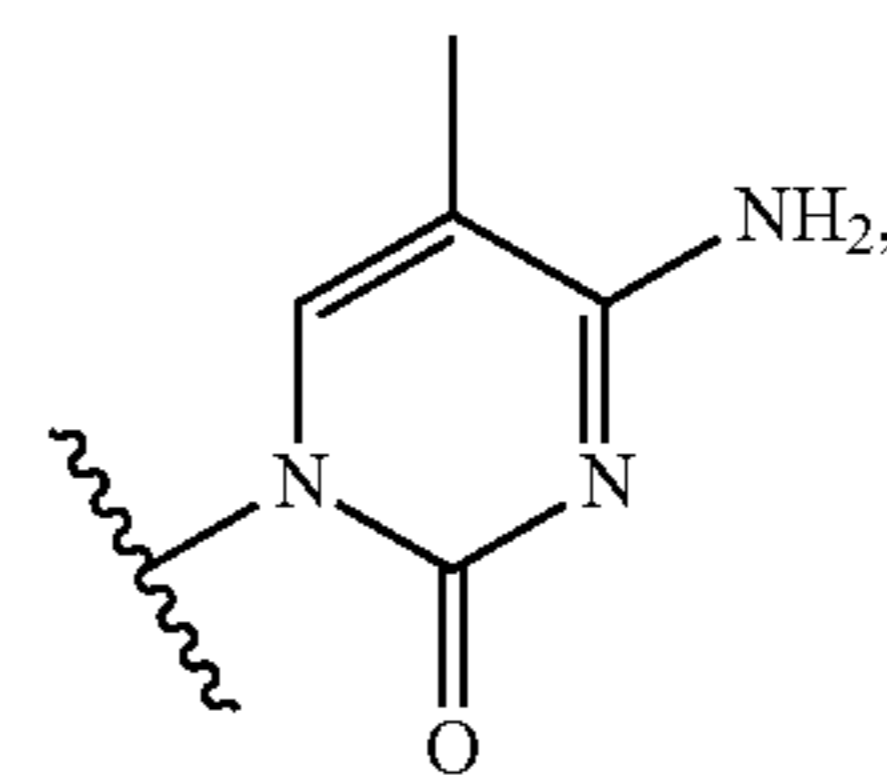
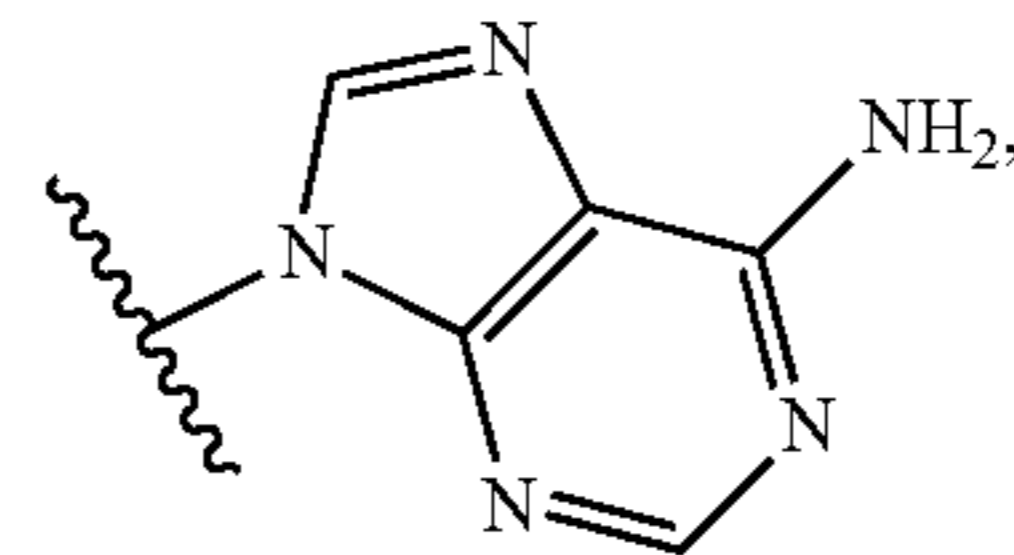
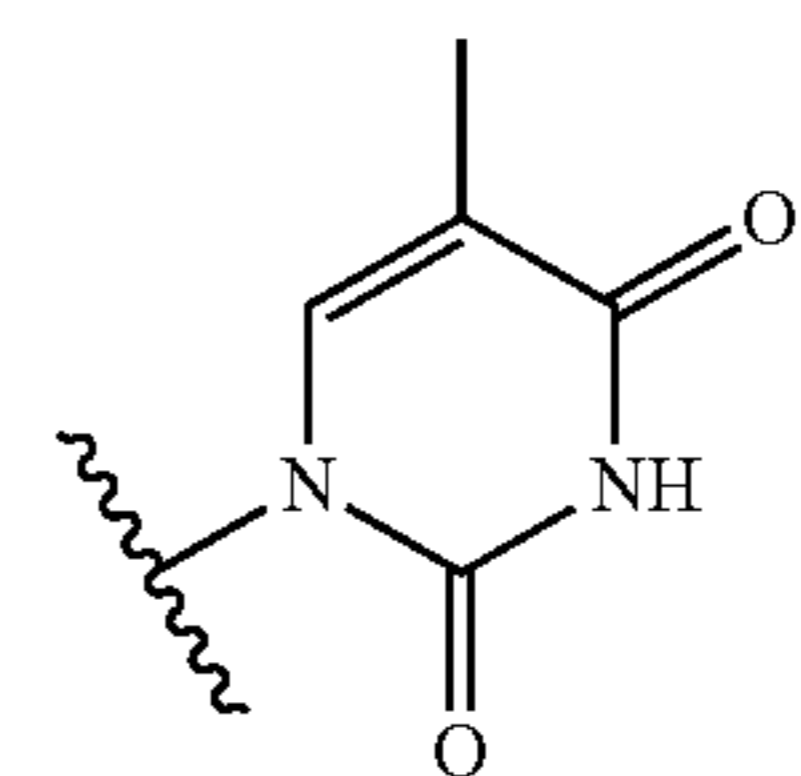
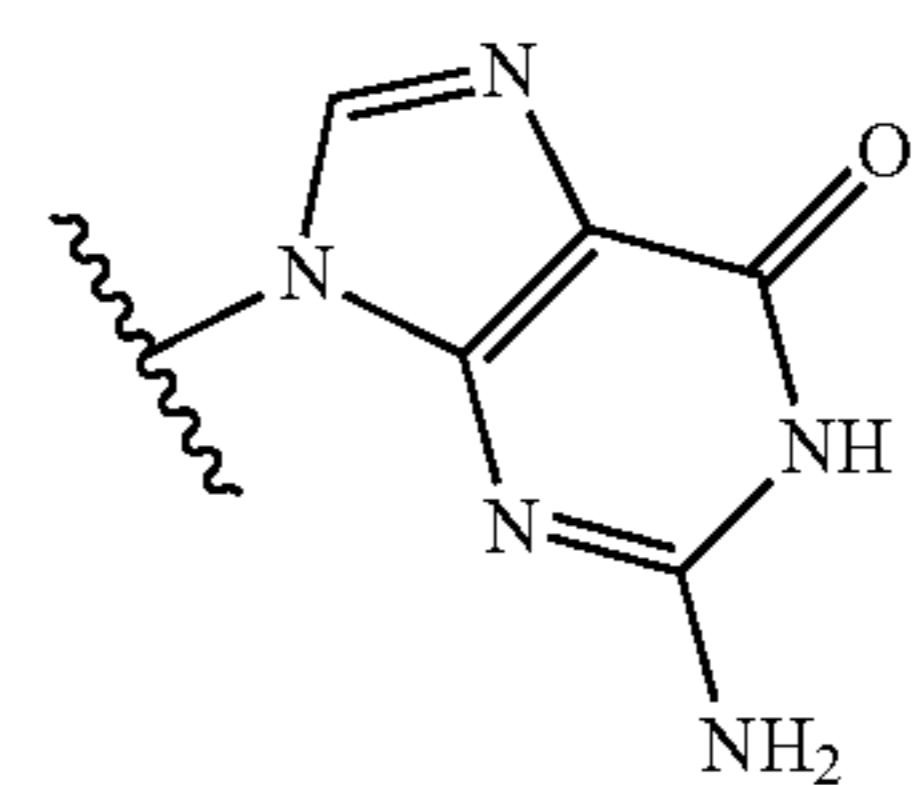
R⁵, R⁶, and R⁷ are each hydrogen, or R⁵ and R⁷ join to form a cycloalkyl ring having from 4 to 7 ring atoms and R⁶ and R⁷ join to form a cycloalkyl ring having from 3 to 4 ring atoms;

R⁴ is, independently at each occurrence, selected from the group consisting of:



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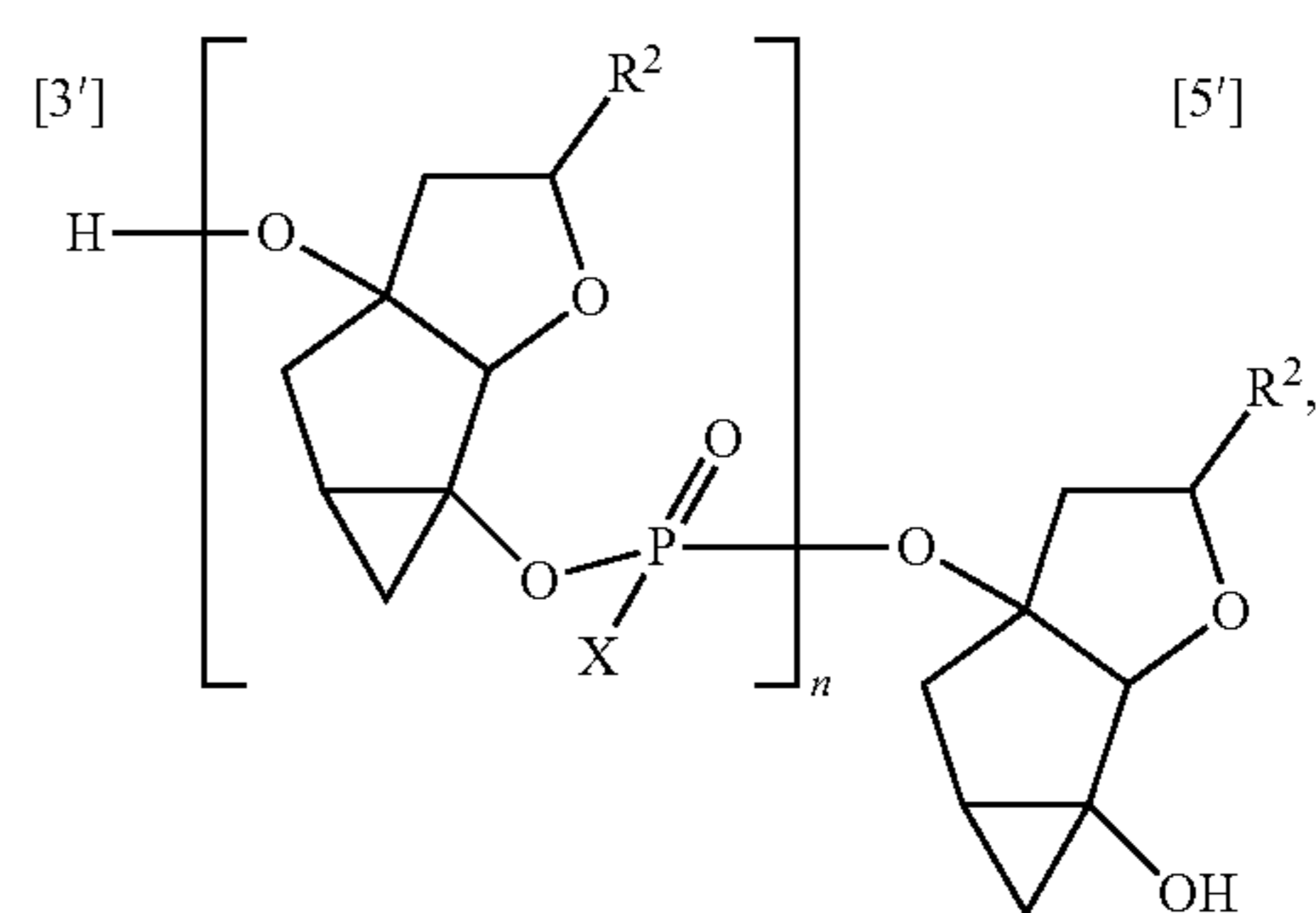
-continued



R⁸ is, independently at each occurrence, selected from the group consisting of H, trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

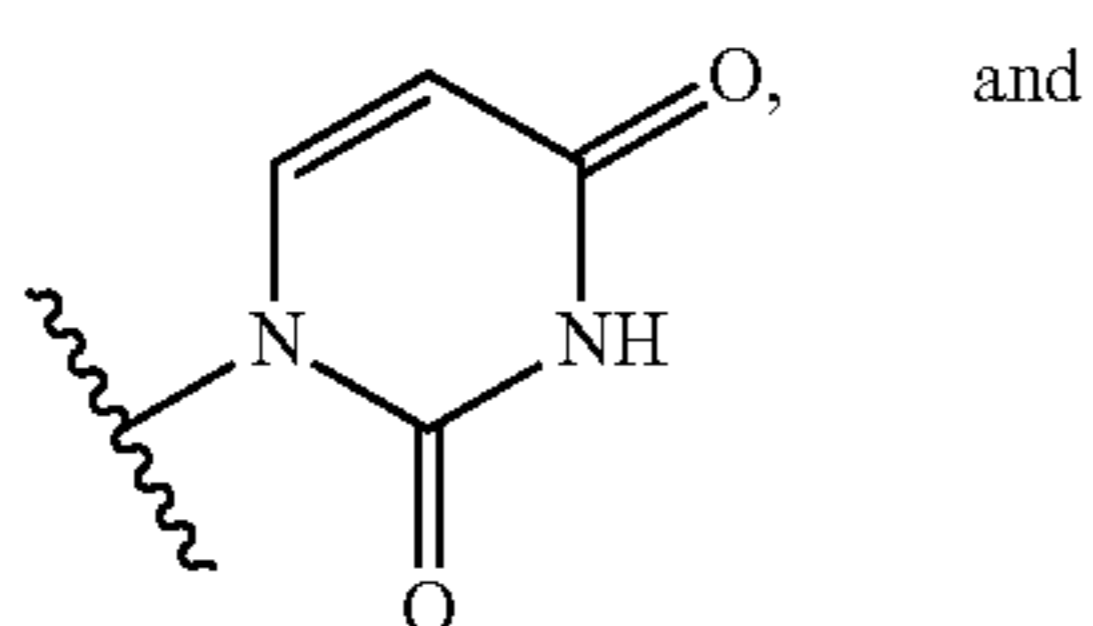
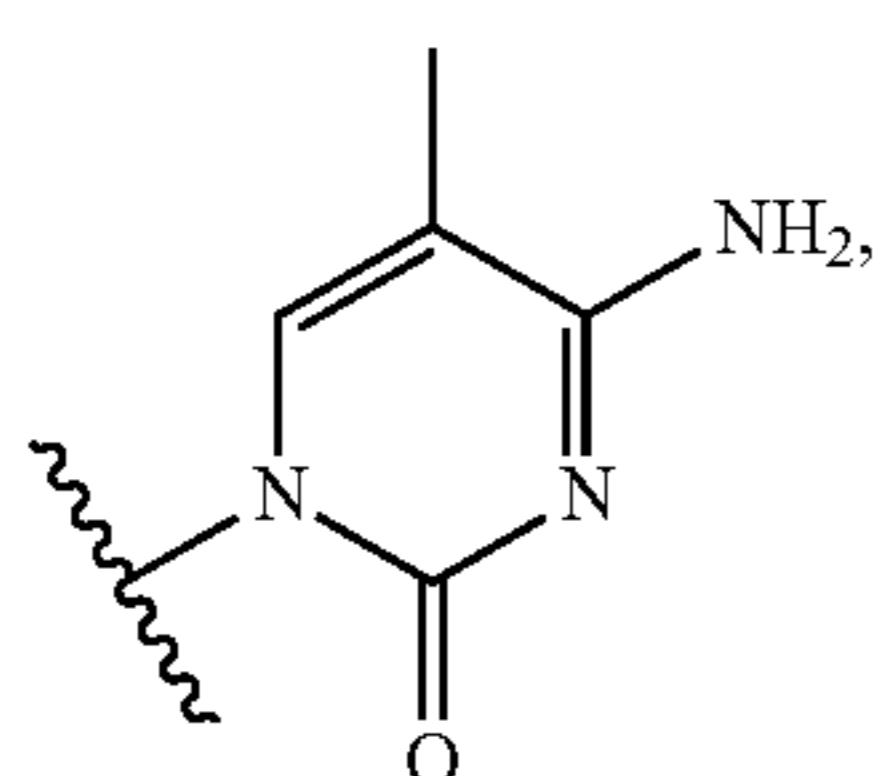
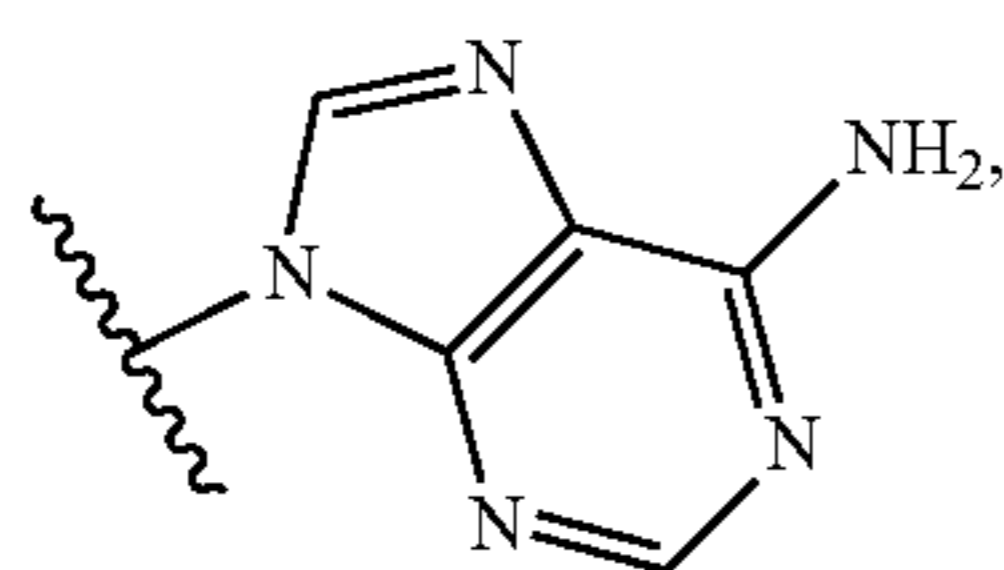
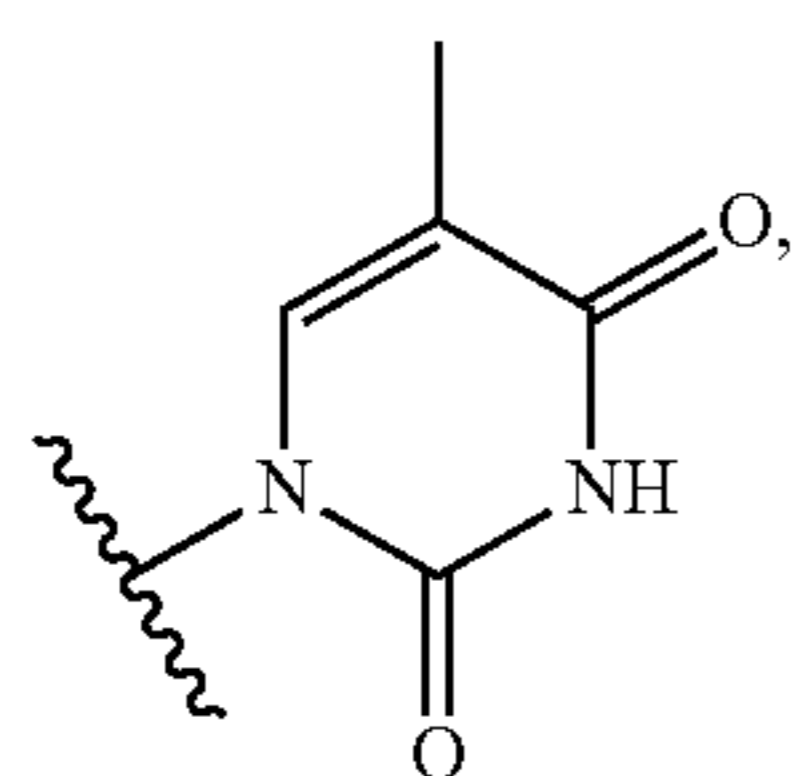
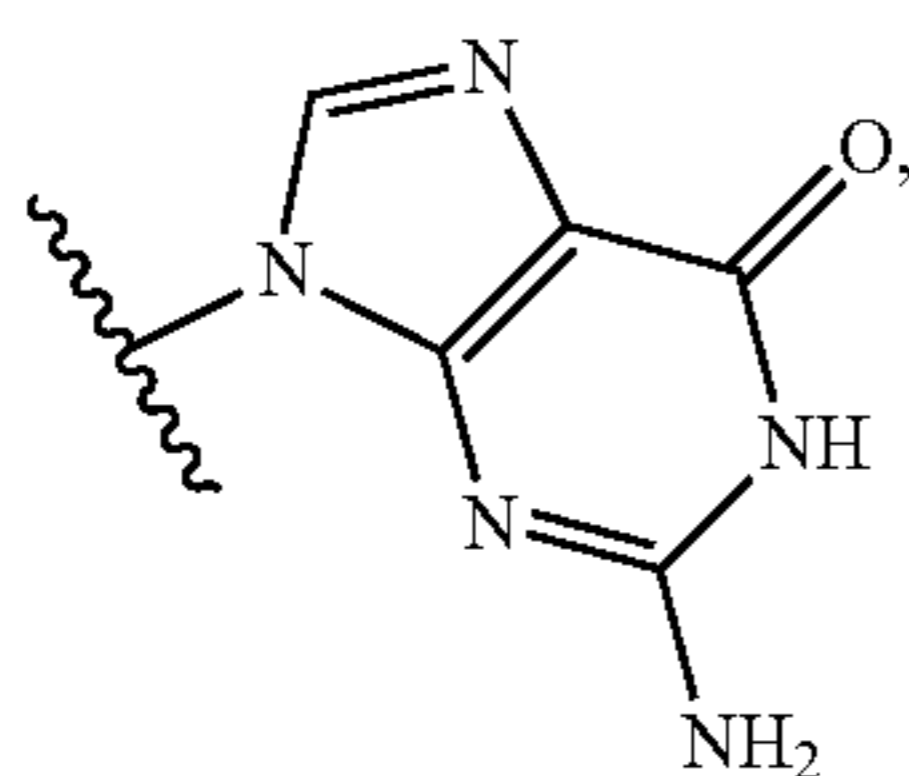
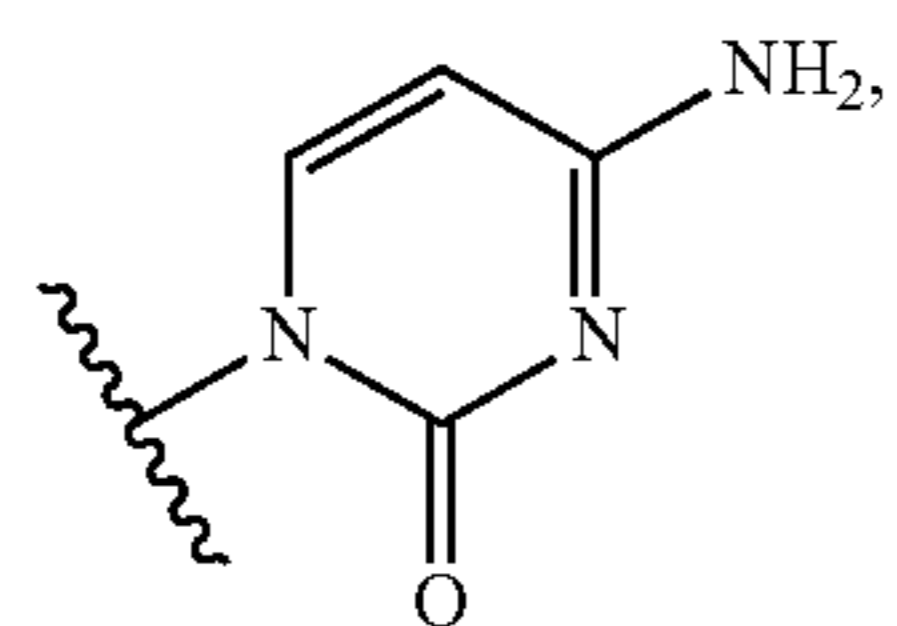
n is 10 to 40; and
X is O or S.

In some embodiments, the oligomeric compound of Formula (D) is of Formula (G):

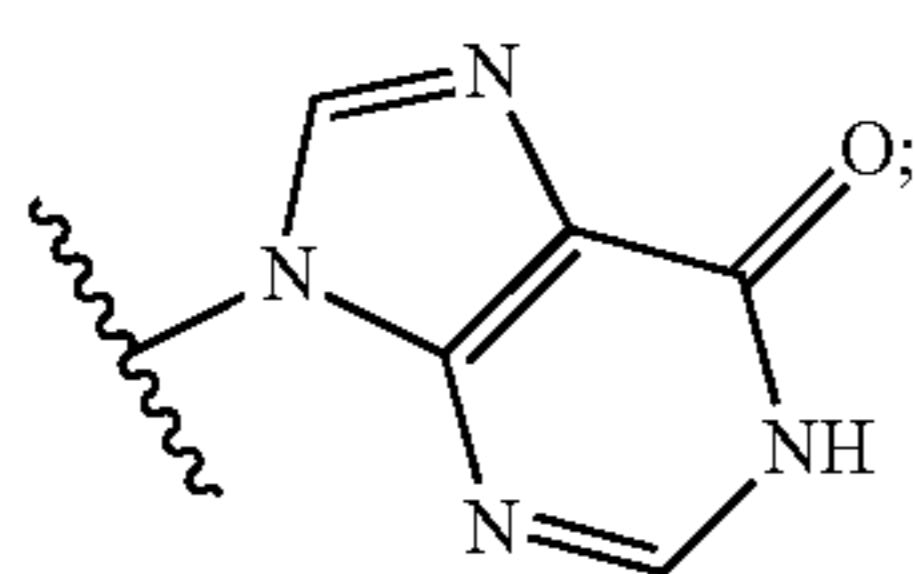


187

wherein R² is, independently at each occurrence, selected from the group consisting of:



and



and

n is 10 to 40.

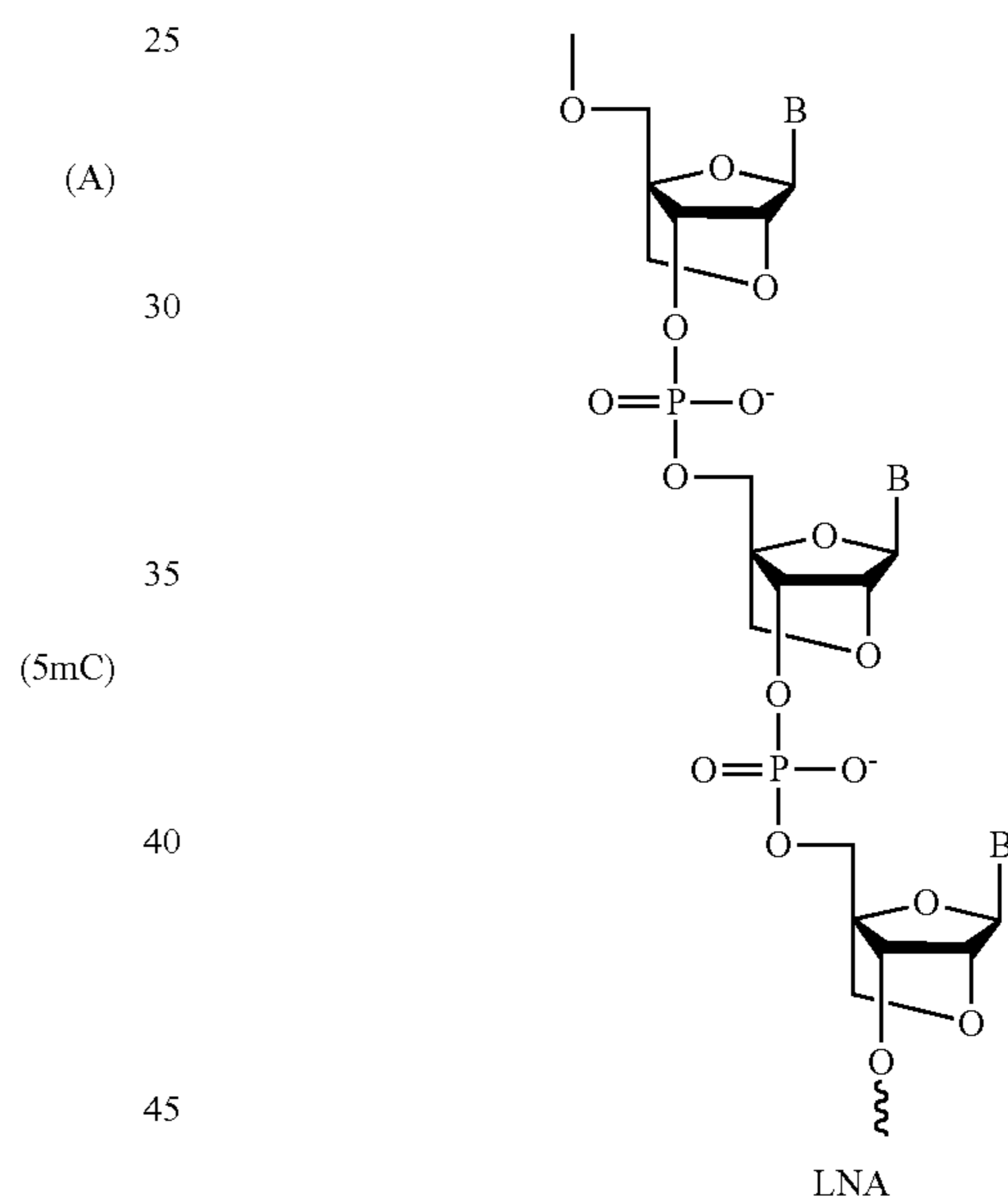
188

Embodiments of the compounds of Formula D and G include, for example:

1. Locked Nucleic Acids (LNAs)

- (C) 5 Modified antisense oligomer compounds may also contain “locked nucleic acid” subunits (LNAs). “LNAs” are a member of a class of modifications called bridged nucleic acid (BNA). BNA is characterized by a covalent linkage that locks the conformation of the ribose ring in a C3'-endo (northern) sugar pucker. For LNA, the bridge is composed of a methylene between the 2'-O and the 4'-C positions. LNA enhances backbone preorganization and base stacking to increase hybridization and thermal stability.

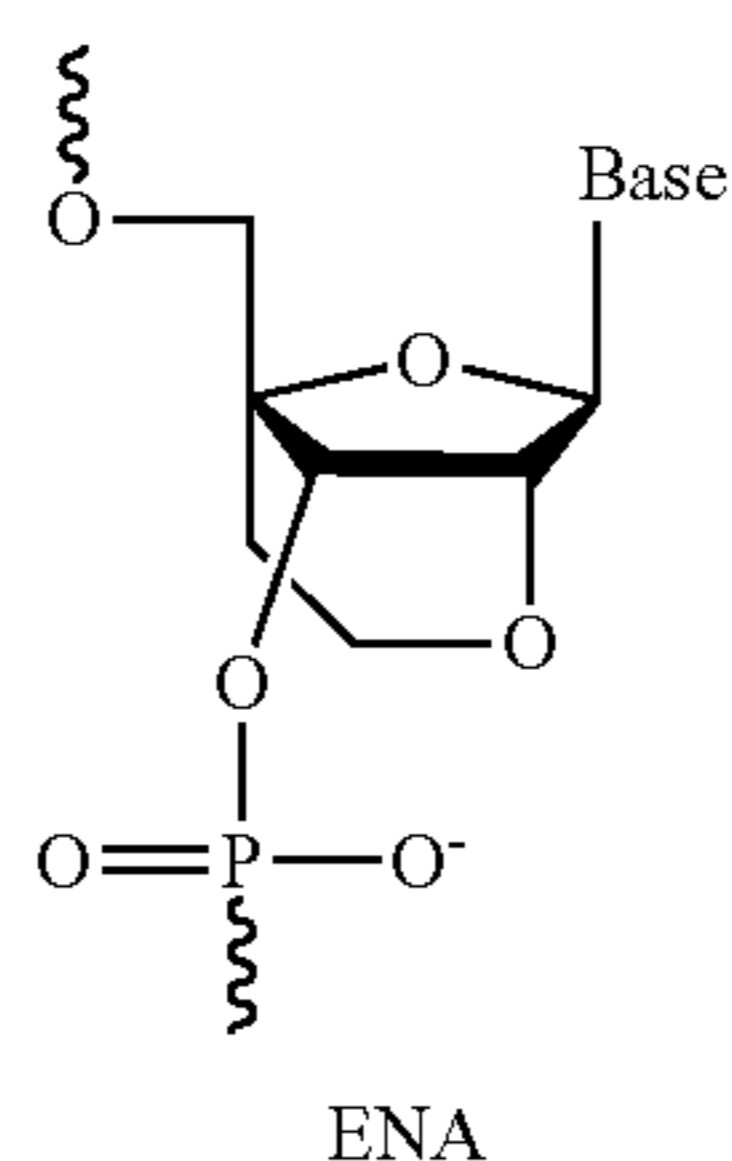
- (G) 10 The structures of LNAs can be found, for example, in Singh et al., *Chem. Commun.* 1998, 455; Wengel, *Tetrahedron* 1998, 54, 3607; Wengel, *Acc. Chem. Res.* 1999, 32, 301; Obika et al., *Tetrahedron Lett.* 1997, 38, 8735; Obika et al., *Tetrahedron Lett.* 1998, 39, 5401; and Obika et al., *Bioorg. Med. Chem.* 2008, 16, 9230, which are hereby incorporated by reference in their entirety. A non-limiting example of an LNA oligomer comprising LNA subunits and phosphodiester internucleoside linkages is depicted below:



- (U) 50 Compounds of the disclosure may incorporate one or more LNAs; in some cases, the compounds may be entirely composed of LNAs. Methods for the synthesis of individual LNA nucleoside subunits and their incorporation into oligomers are described, for example, in U.S. Pat. Nos. 7,572, 582, 7,569,575, 7,084,125, 7,060,809, 7,053,207, 7,034,133, 6,794,499, and 6,670,461, which are hereby incorporated by reference in their entirety. Typical internucleoside linkers include phosphodiester and phosphorothioate moieties; alternatively, non-phosphorous containing linkers may be employed. Further embodiments include an LNA containing compound where each LNA subunit is separated by a DNA subunit. Certain compounds are composed of alternating LNA and DNA subunits where the internucleoside linker is phosphorothioate.

- (I) 60 2'-O,4'-C-ethylene-bridged nucleic acids (ENAs) are another member of the class of BNAs. A non-limiting example of an ENA subunit and phosphodiester internucleoside linkage is depicted below:

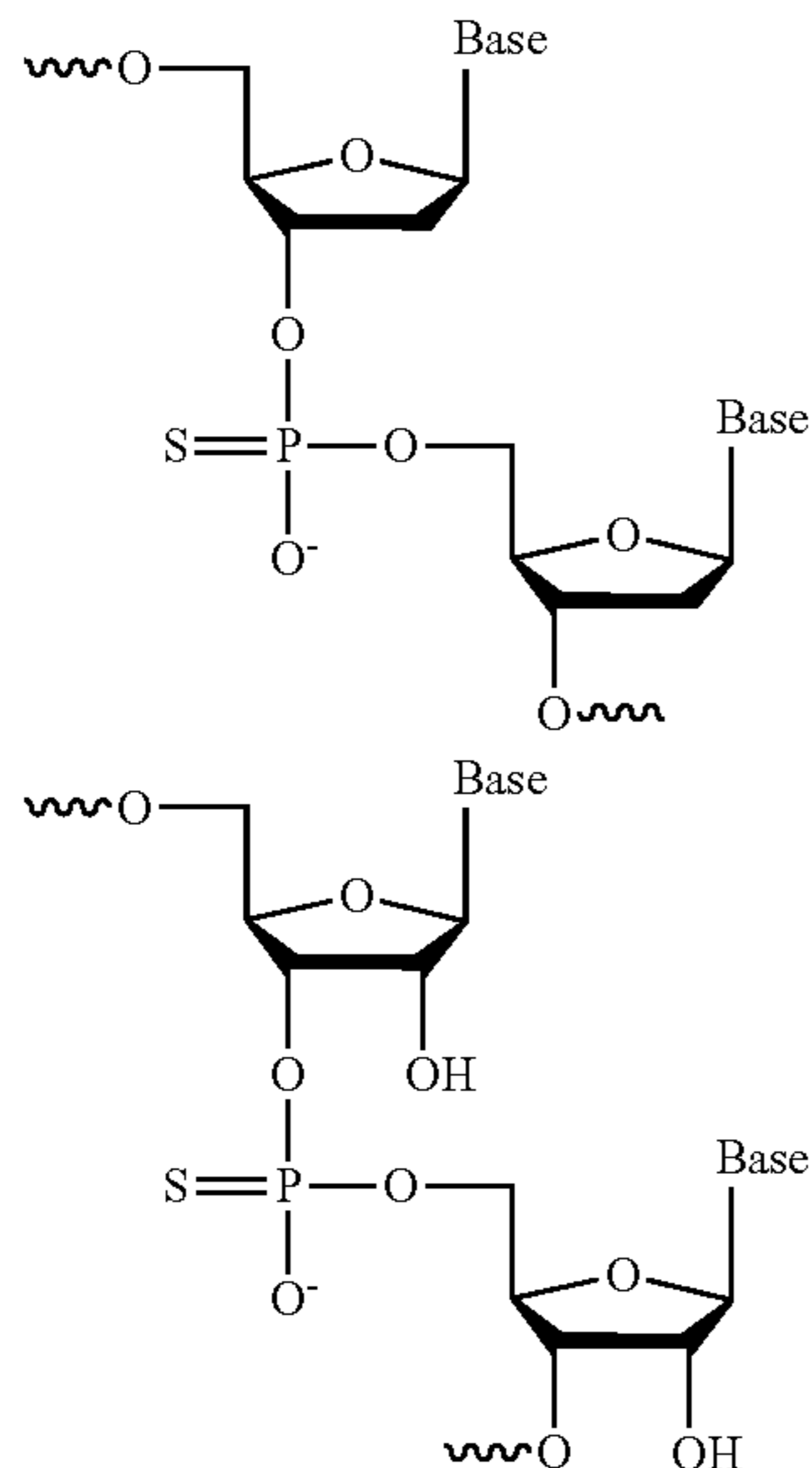
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ENA oligomers and their preparation are described in Obika et al., *Tetrahedron Lett.* 1997, 38, 8735, which is hereby incorporated by reference in its entirety. Compounds of the disclosure may incorporate one or more ENA sub-

2. Phosphorothioates

“Phosphorothioates” (or S-oligos) are a variant of native DNA or RNA in which one of the nonbridging oxygens of the phosphodiester internucleoside linkages is replaced by sulfur. A non-limiting example of a phosphorothioate DNA (left), comprising deoxyribose subunits and phosphorothioate internucleoside linkages, and phosphorothioate RNA (right), comprising ribose subunits and phosphorothioate internucleoside linkages, are depicted below:



The sulfurization of the internucleoside bond reduces the action of endo- and exonucleases including 5' to 3' and 3' to 5' DNA Pol 1 exonuclease, nucleases S and P1, RNases, serum nucleases and snake venom phosphodiesterase. Phosphorothioates may be made by two principal routes: by the action of a solution of elemental sulfur in carbon disulfide on a hydrogen phosphonate, or by the method of sulfurizing phosphite triesters with either tetraethylthiuram disulfide (TETD) or 3H-1, 2-bensodithiol-3-one 1, 1-dioxide (BDTD) (see, e.g., Iyer et al., *J. Org. Chem.* 1990, 55, 4693-4699, which is hereby incorporated by reference in its entirety). The latter methods avoid the problem of elemental sulfur's insolubility in most organic solvents and the toxicity of

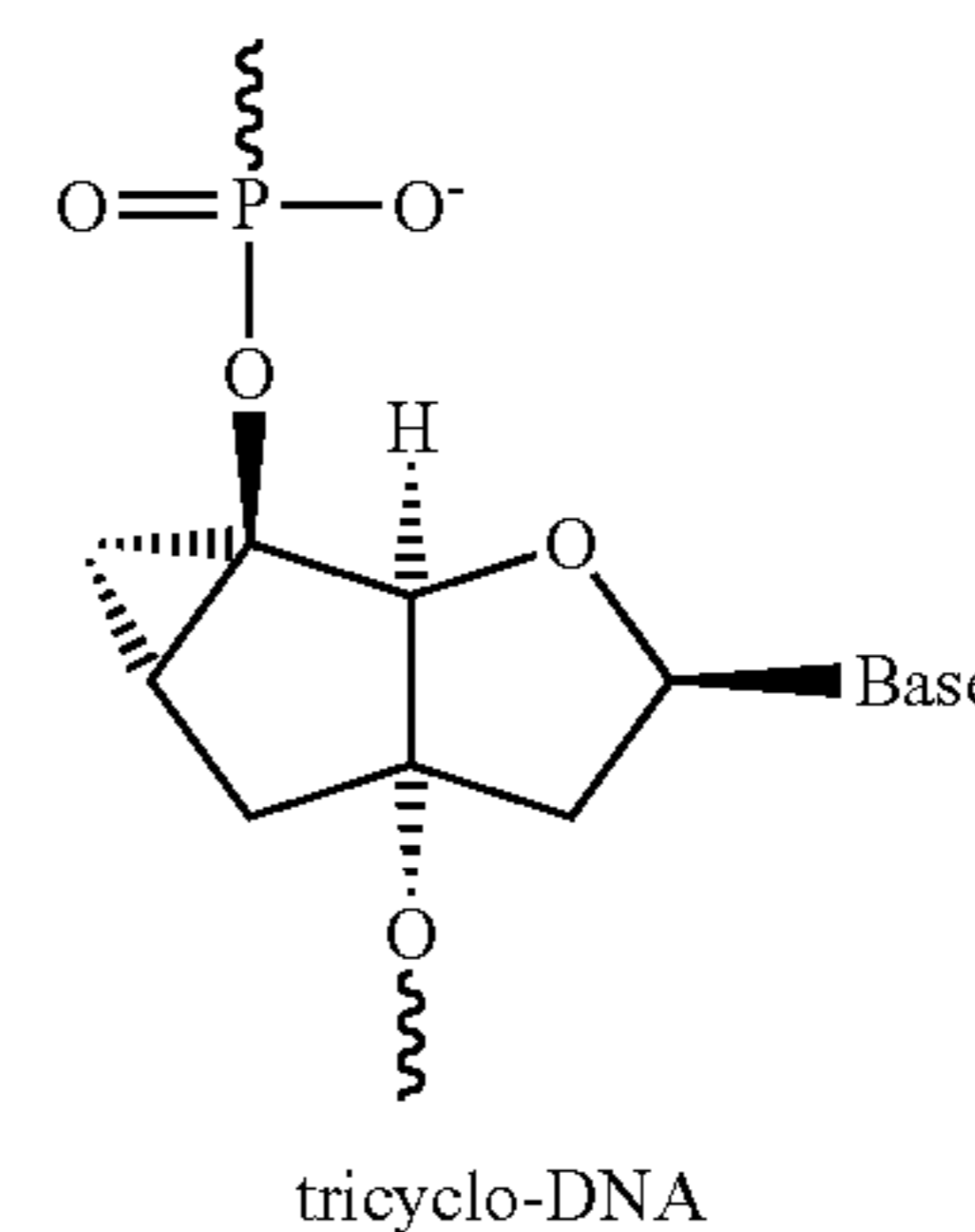
190

carbon disulfide. The TETD and BDTD methods also yield higher purity phosphorothioates.

3. Tricyclo-DNAs and Tricyclo-Phosphorothioate Nucleotides

Tricyclo-DNAs (tc-DNA) are a class of constrained DNA analogs in which each nucleotide is modified by the introduction of a cyclopropane ring to restrict conformational flexibility of the backbone and to optimize the backbone geometry of the torsion angle γ . Homobasic adenine- and thymine-containing tc-DNAs form extraordinarily stable A-T base pairs with complementary RNAs. Tricyclo-DNAs and their synthesis are described in PCT Publication No. WO 2010/115993, which is hereby incorporated by reference in its entirety. Compounds of the disclosure may incorporate one or more tricyclo-DNA subunits; in some cases, the compounds may be entirely composed of tricyclo-DNA subunits.

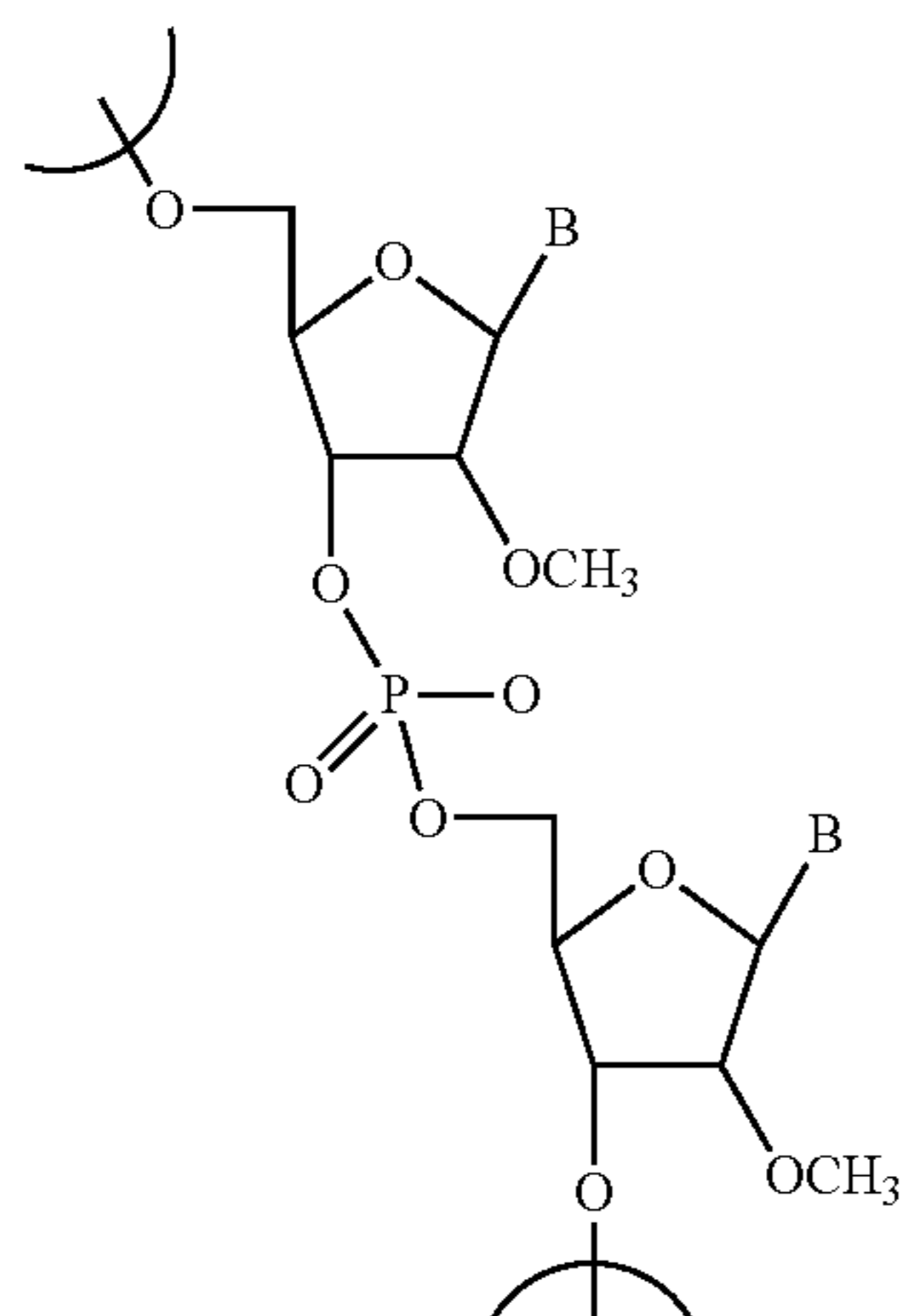
Tricyclo-phosphorothioate nucleotides are tricyclo-DNA subunits with phosphorothioate internucleoside linkages. Tricyclo-phosphorothioate nucleotides and their synthesis are described in PCT Publication No. WO 2013/053928, which is hereby incorporated by reference in its entirety. Compounds of the disclosure may incorporate one or more tricyclo-DNA subunits; in some cases, the compounds may be entirely composed of tricyclo-DNA nucleotides. A non-limiting example of a tricyclo-DNA/tricyclo subunit and phosphodiester internucleoside linkage is depicted below:



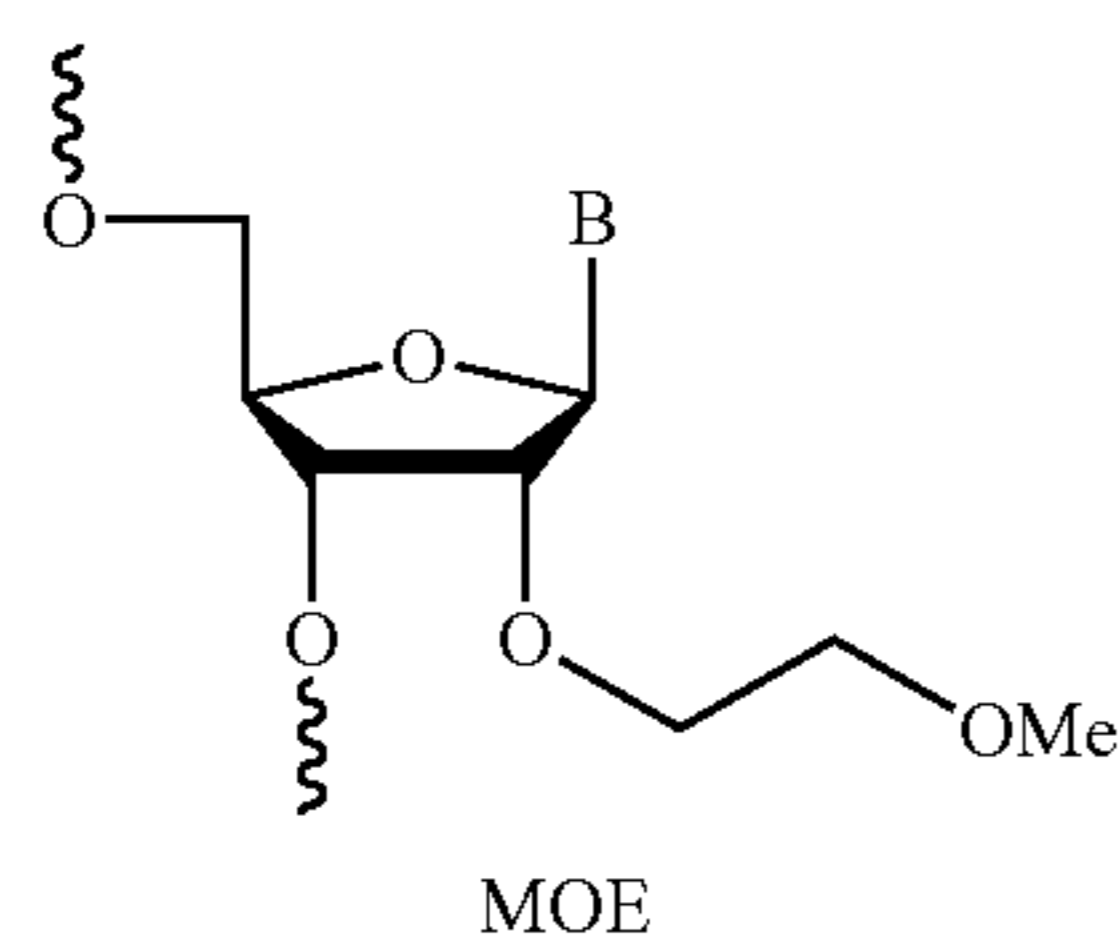
4. 2' O-Methyl, 2' O-MOE, and 2'-F Oligomers

“2'O-Me oligomer” molecules comprise subunits that carry a methyl group at the 2'-OH residue of the ribose molecule. 2'-O-Me-RNAs show the same (or similar) behavior as DNA, but are protected against nuclease degradation. 2'-O-Me-RNAs can also be combined with phosphorothioate oligomers (PTOs) for further stabilization. 2'O-Me oligomers (wherein the 2'-OMe subunits are connected by phosphodiester or phosphorothioate internucleoside linkages) can be synthesized according to routine techniques in the art (see, e.g., Yoo et al., *Nucleic Acids Res.* 2004, 32, 2008-16, which is hereby incorporated by reference in its entirety). A non-limiting example of a 2' O-Me oligomer comprising 2'-OMe subunits and phosphodiester intersubunit linkages is depicted below:

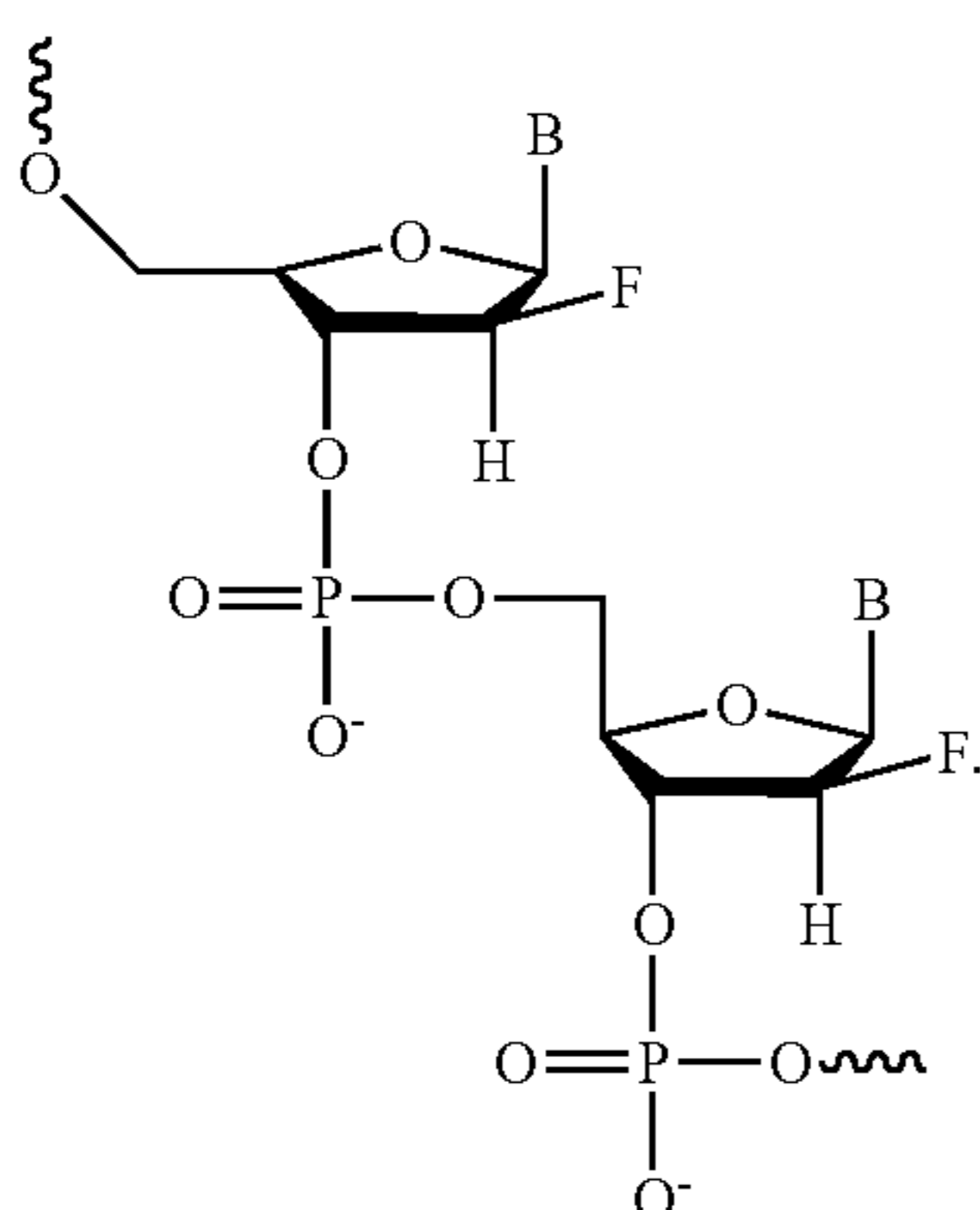
191



2' O-Me oligomers may also comprise a phosphorothioate linkage (2' O-Me phosphorothioate oligomers). 2' O-Methoxyethyl oligomers (2' O-MOE), like 2' O-Me oligomers, comprise subunits that carry a methoxyethyl group at the 2'-OH residue of the ribose molecule and are discussed in Martin et al., *Helv. Chim. Acta* 1995, 78, 486-504, which is hereby incorporated by reference in its entirety. A non-limiting example of a 2' O-MOE subunit is depicted below:



In contrast to the preceding alkylated 2'OH ribose derivatives, 2'-fluoro oligomers comprise subunits that have a fluoro radical at the 2' position in place of the 2'OH. A non-limiting example of a 2'-F oligomer comprising 2'-F subunits and phosphodiester internucleoside linkages is depicted below:



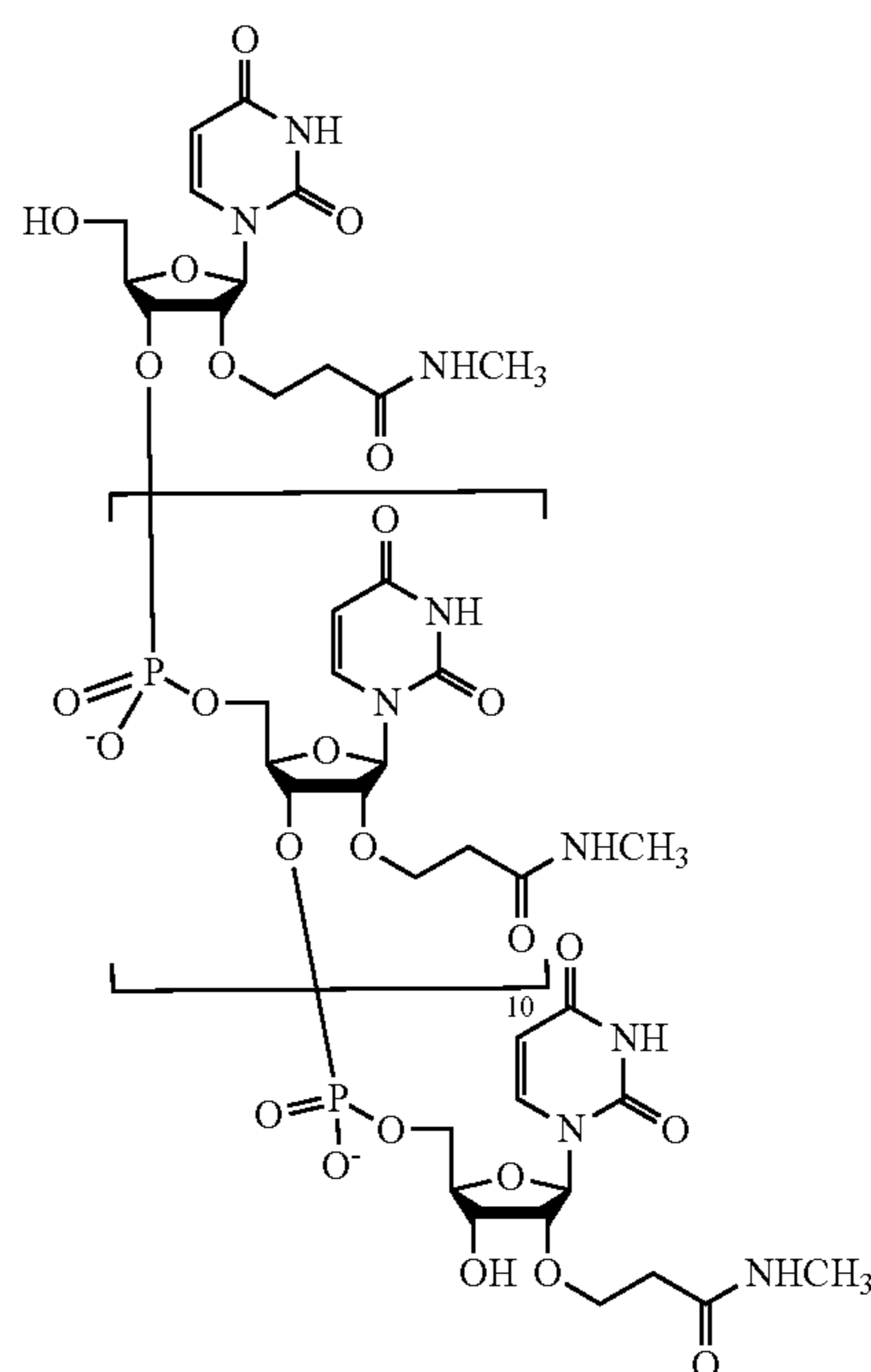
2'-fluoro oligomers are further described in WO 2004/043977, which is hereby incorporated by reference in its entirety. Compounds of the disclosure may incorporate one or more 2'-O-Methyl, 2' O-MOE, and 2'-F subunits and may

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utilize any of the internucleoside linkages described here. In some instances, a compound of the disclosure could be composed of entirely 2'-O-Methyl, 2' O-MOE, or 2'-F subunits. One embodiment of a compound of the disclosure is composed entirely of 2'-O-methyl subunits.

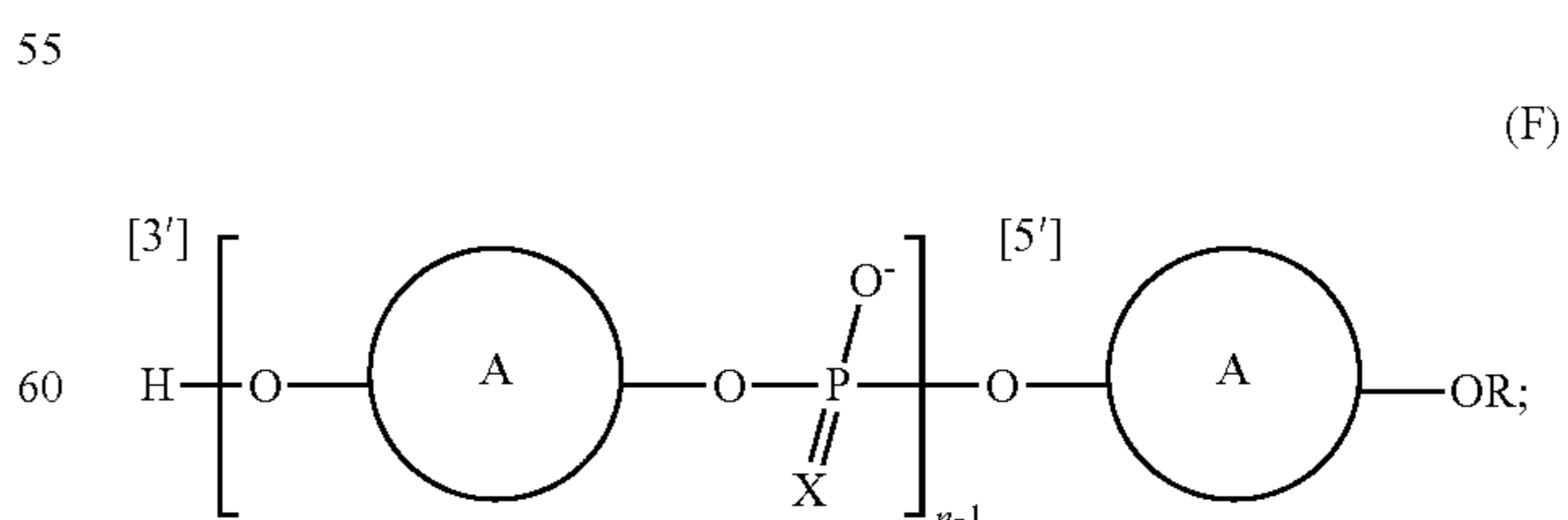
5. 2'-O-[2-(N-methylcarbamoyl)ethyl] Oligomers (MCEs)

MCEs are another example of 2'O modified ribonucleotides useful in the compounds of the disclosure. Here, the 2'OH is derivatized to a 2-(N-methylcarbamoyl)ethyl moiety to increase nuclease resistance. A non-limiting example of an MCE oligomer comprising MCE subunits and phosphodiester internucleoside linkages is depicted below:



MCEs and their synthesis are described in Yamada et al., *J. Org. Chem.* 2011, 76(9), 3042-53, which is hereby incorporated by reference in its entirety. Compounds of the disclosure may incorporate one or more MCE subunits.

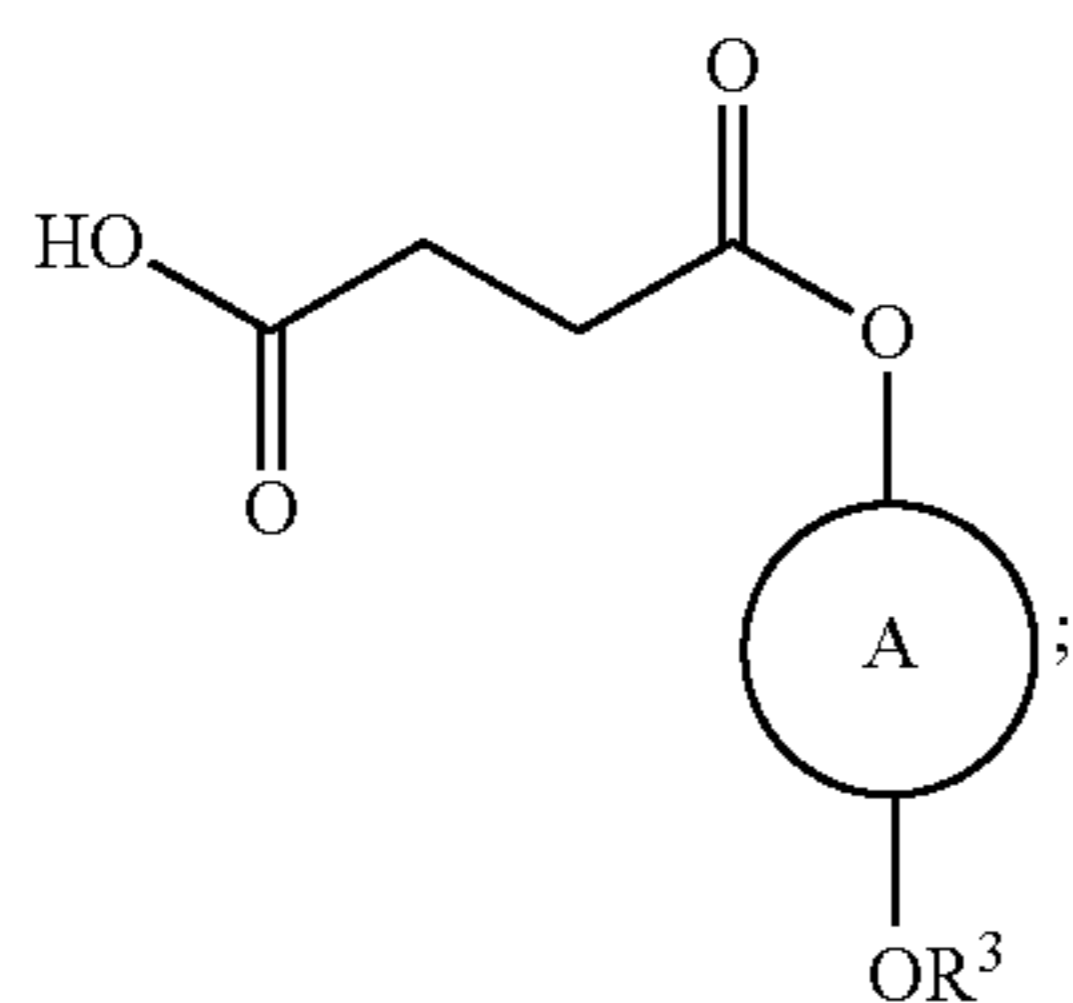
In another aspect, provided herein is a process for preparing an oligomeric compound of Formula (F):



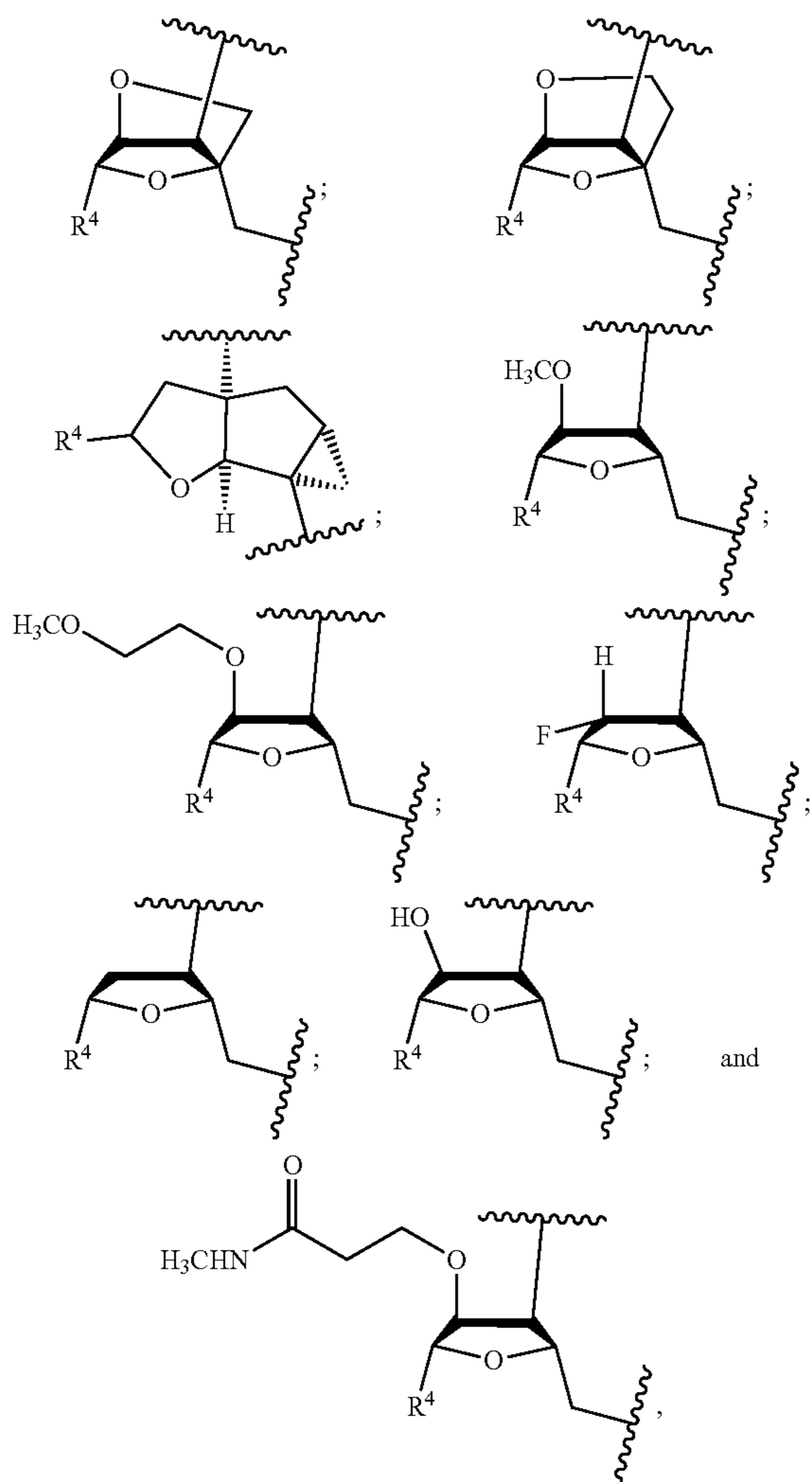
wherein:

each A is, independently for each occurrence, selected from the group consisting of:

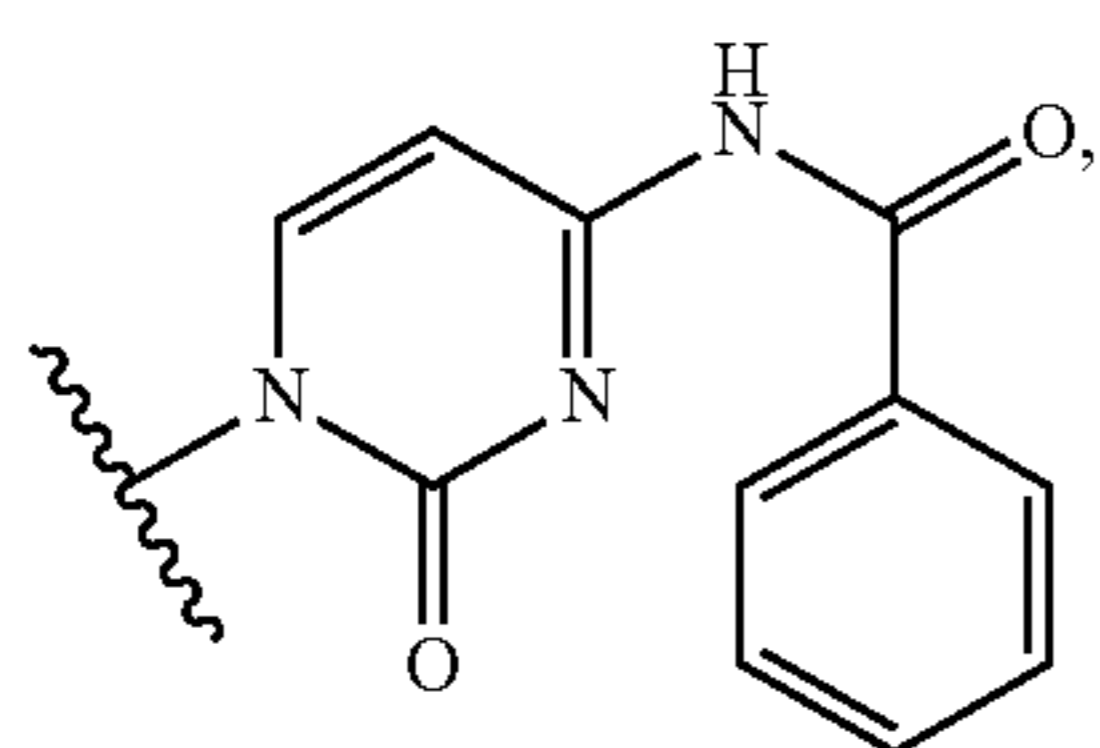
195



wherein A is selected from the group consisting of:



wherein R⁴ is selected from the group consisting of:



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-continued

(F2)

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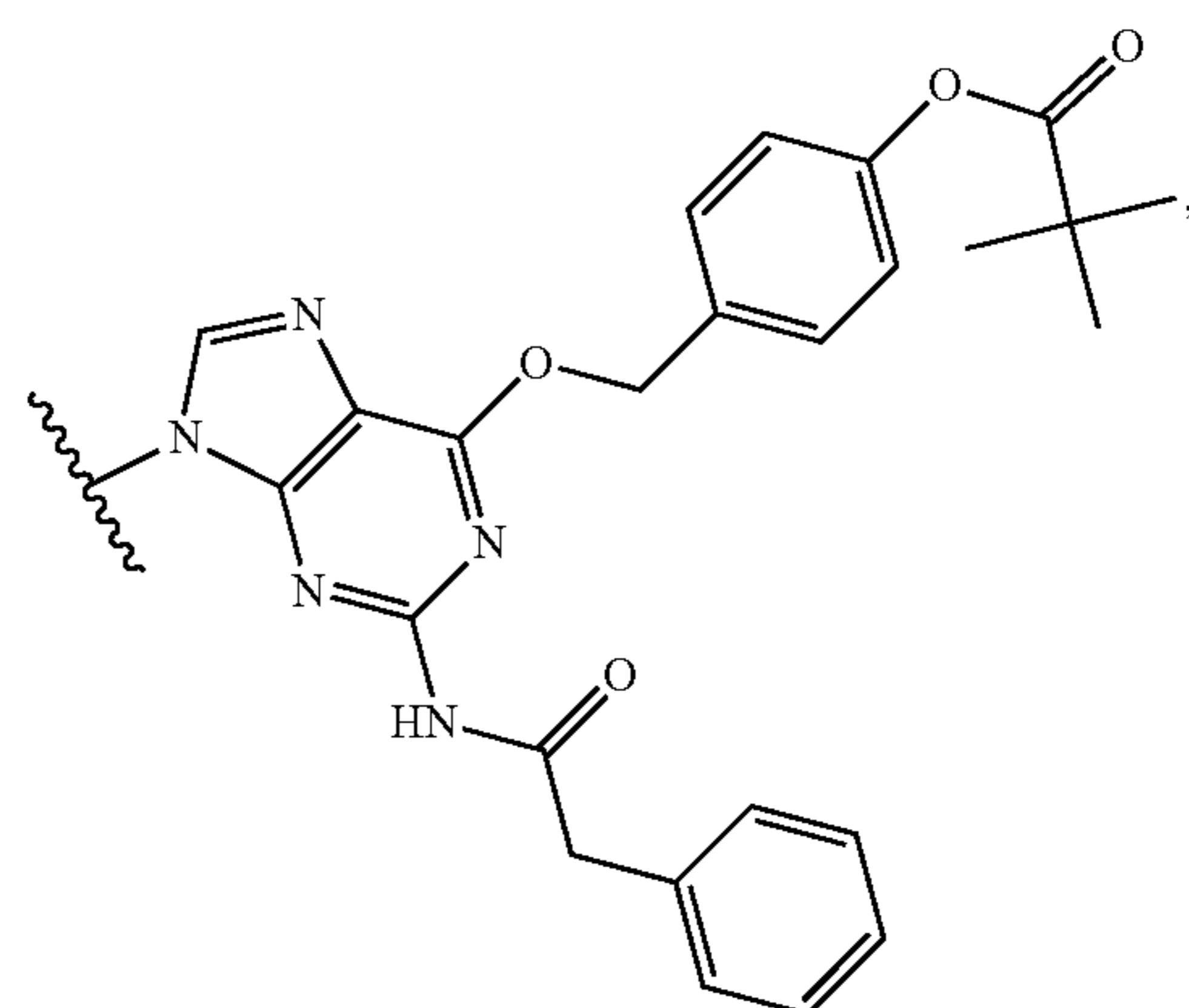
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(PC)

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(DPG)

(T)

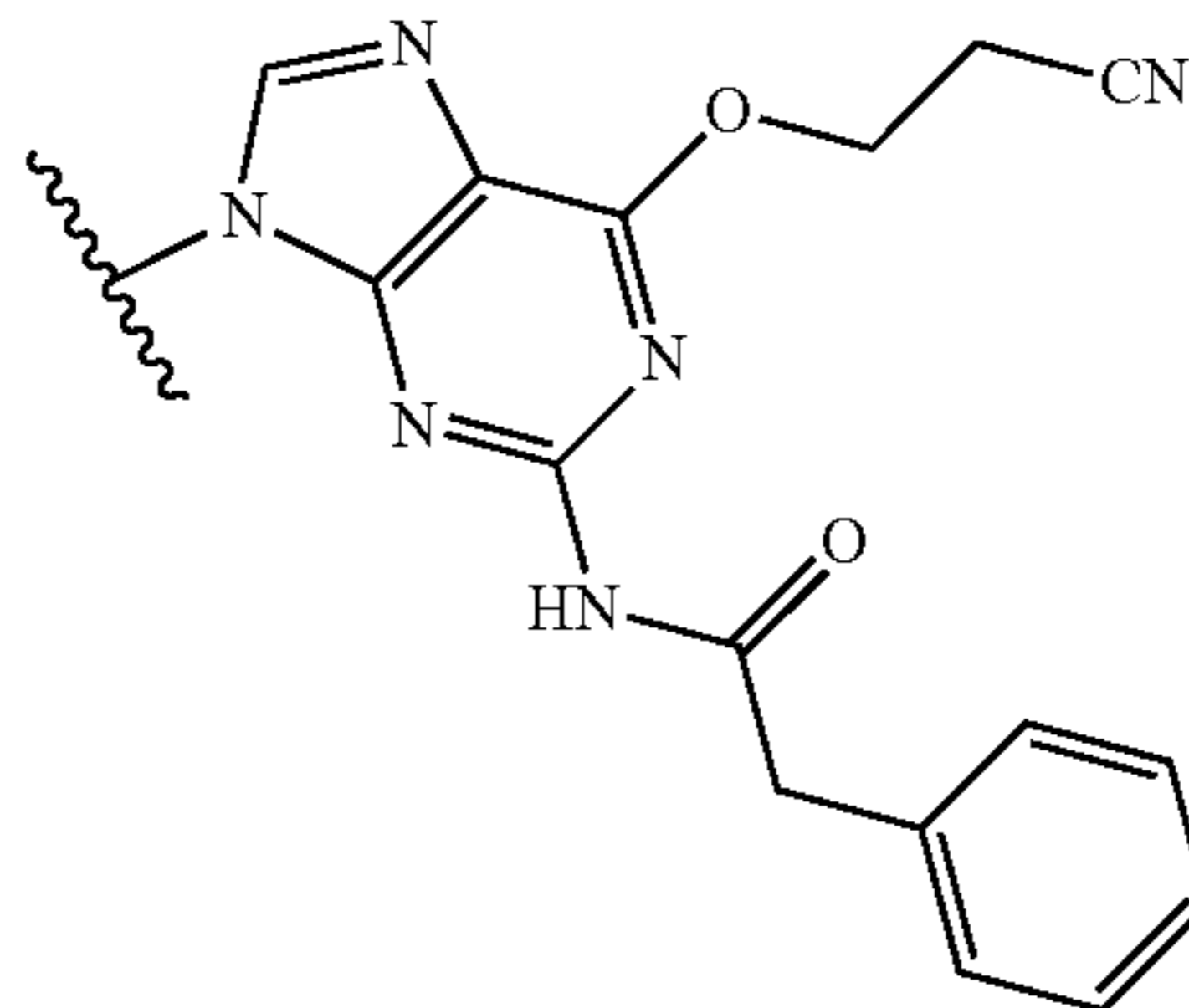
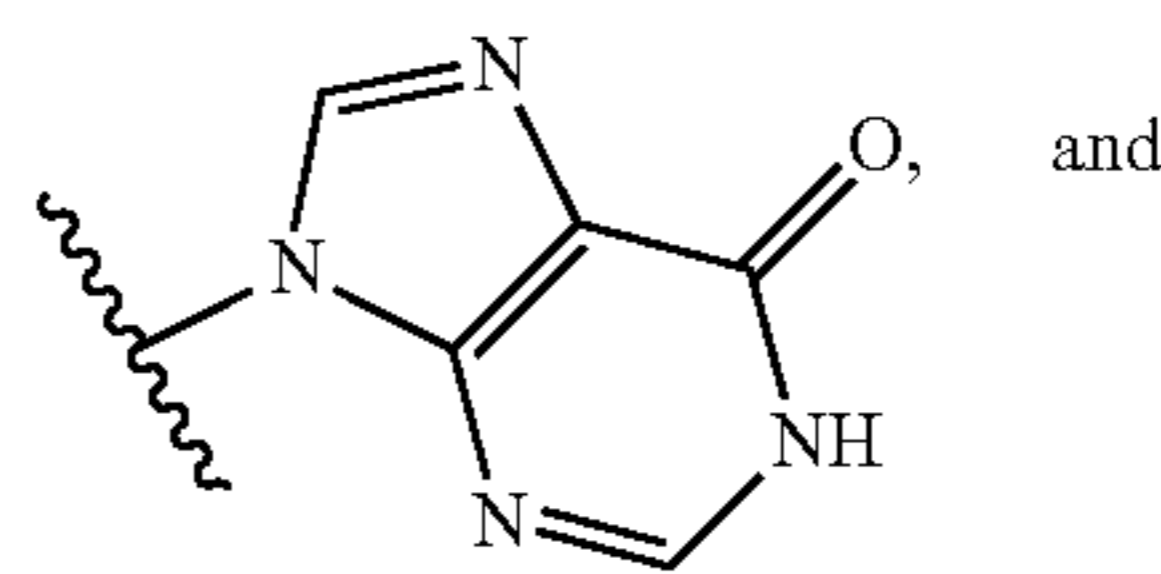
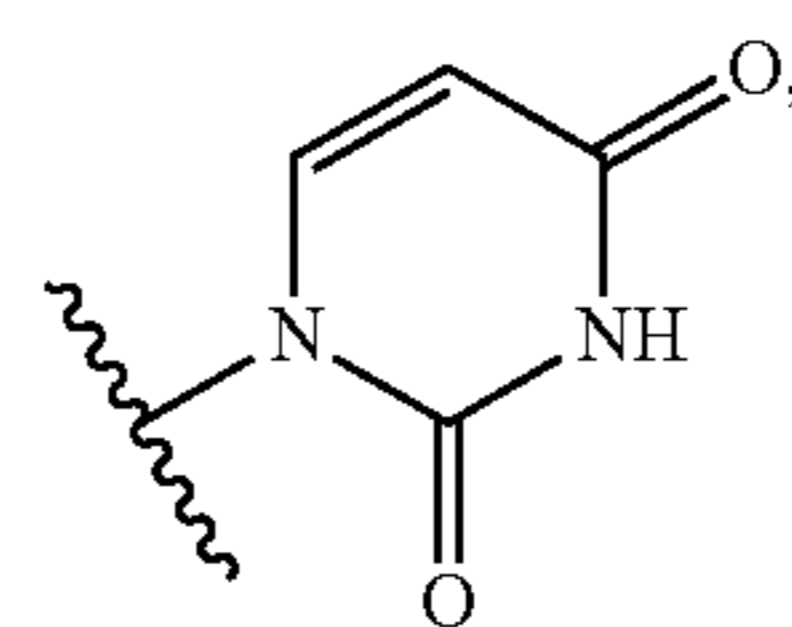
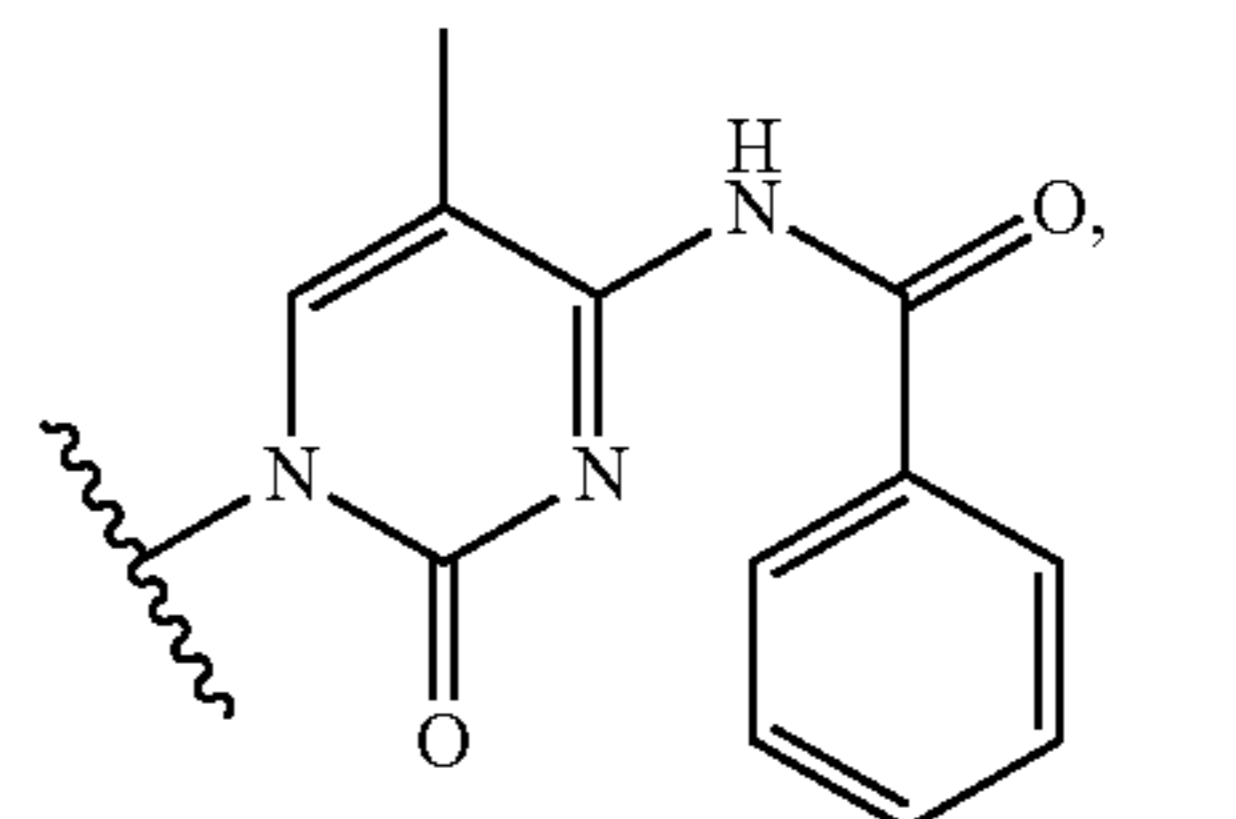
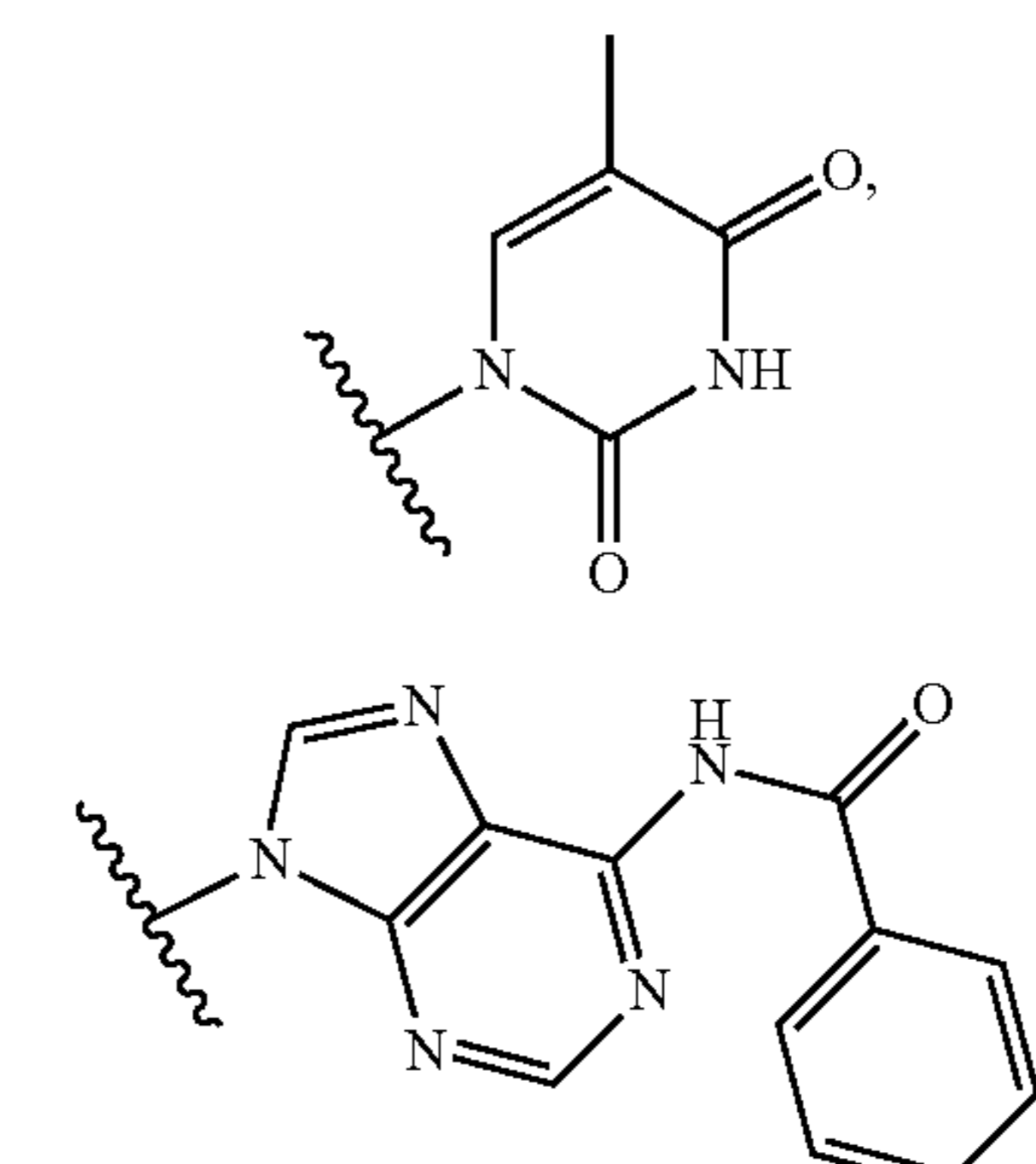
(PA)

(P5mC)

(U)

(I)

(PG)

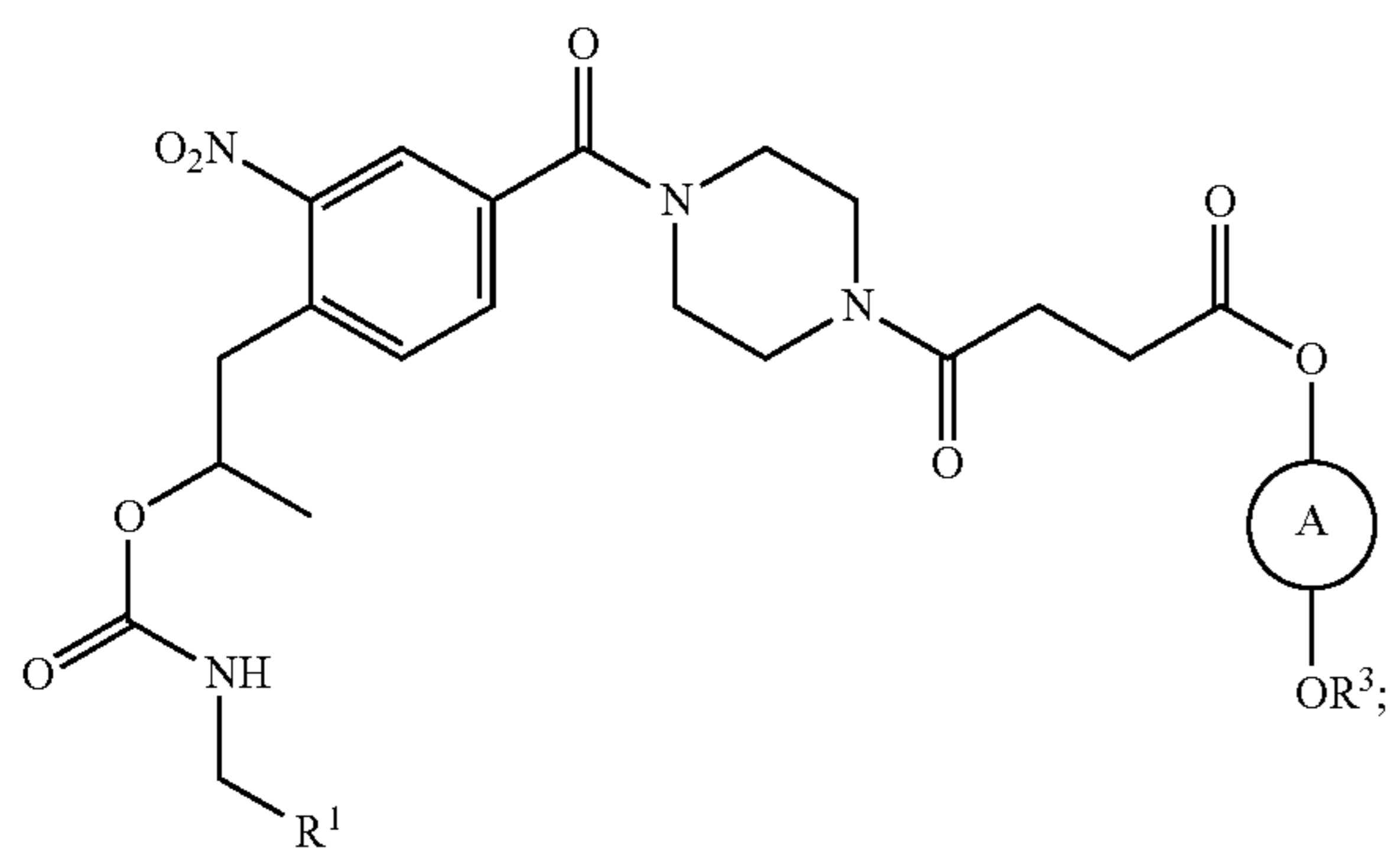


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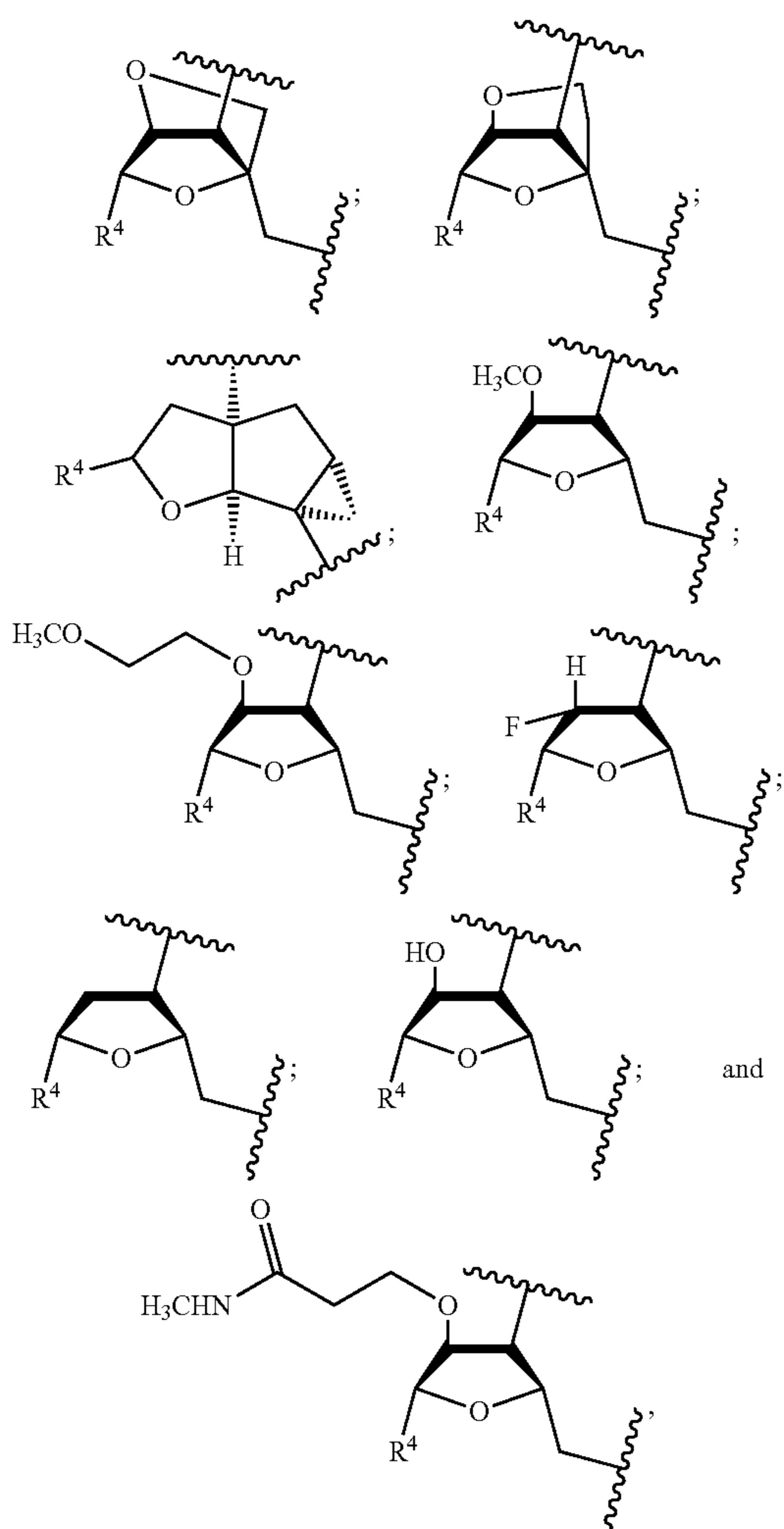
197

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl

and trimethoxytrityl, and PG is a protecting group; to form a compound of Formula (F3):

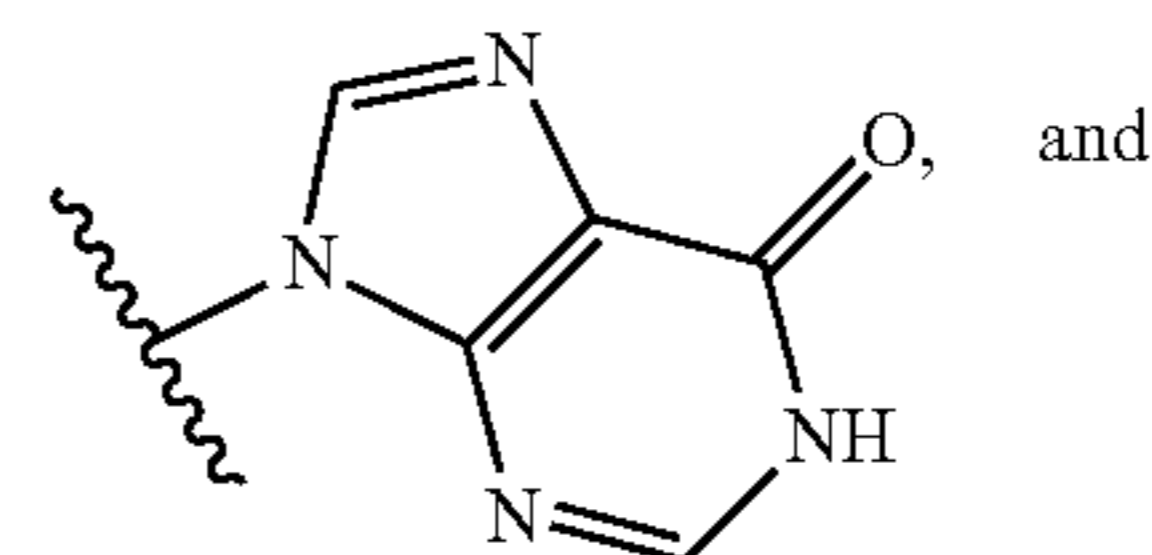
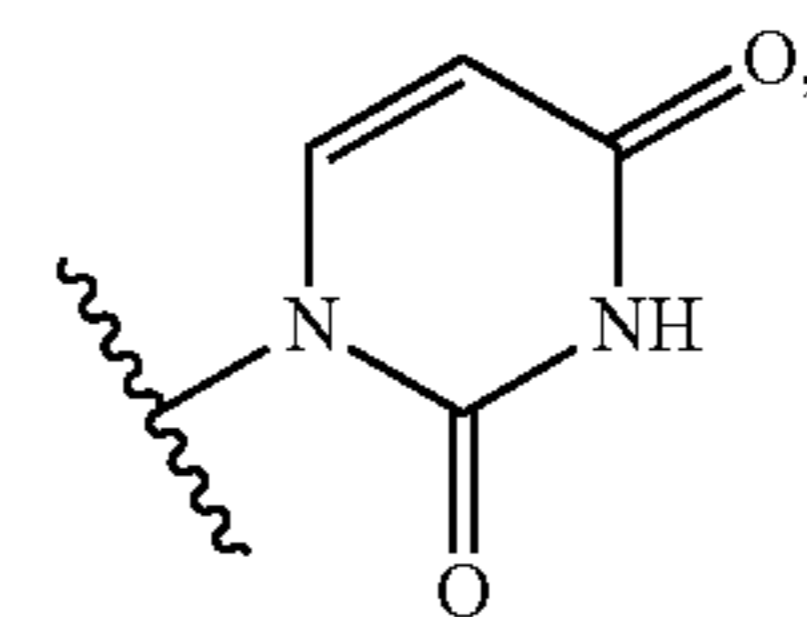
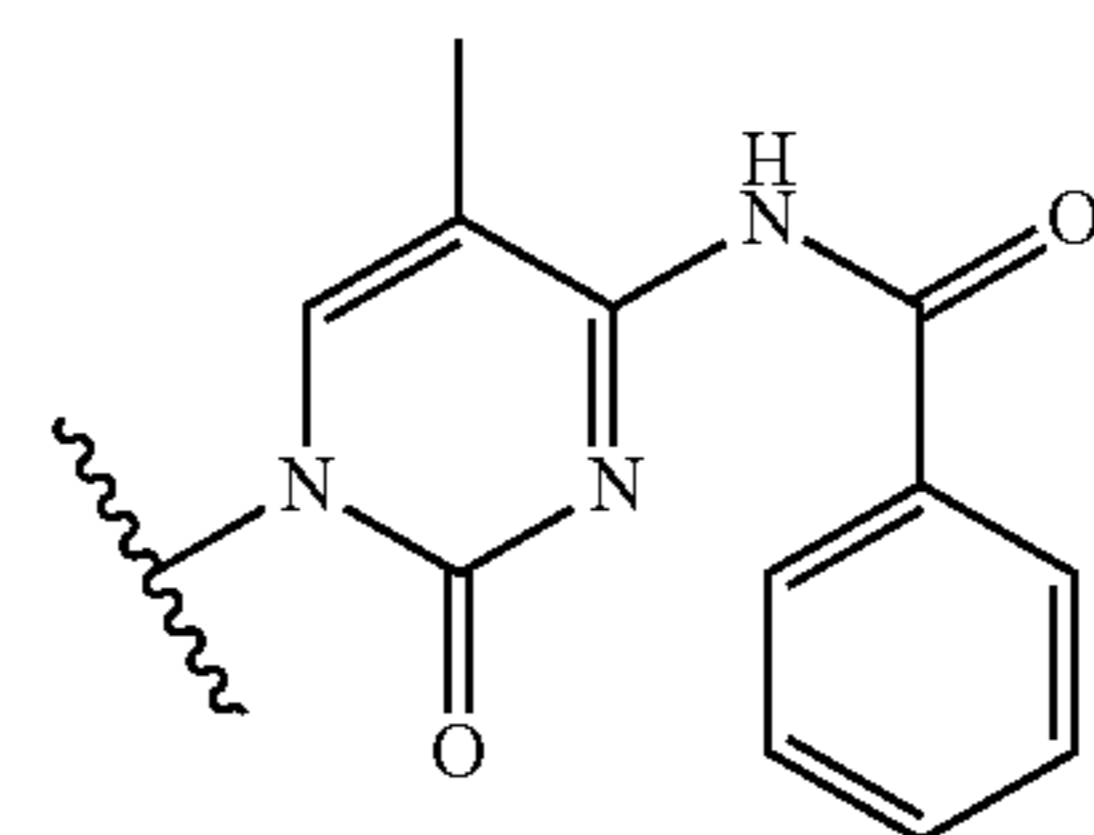
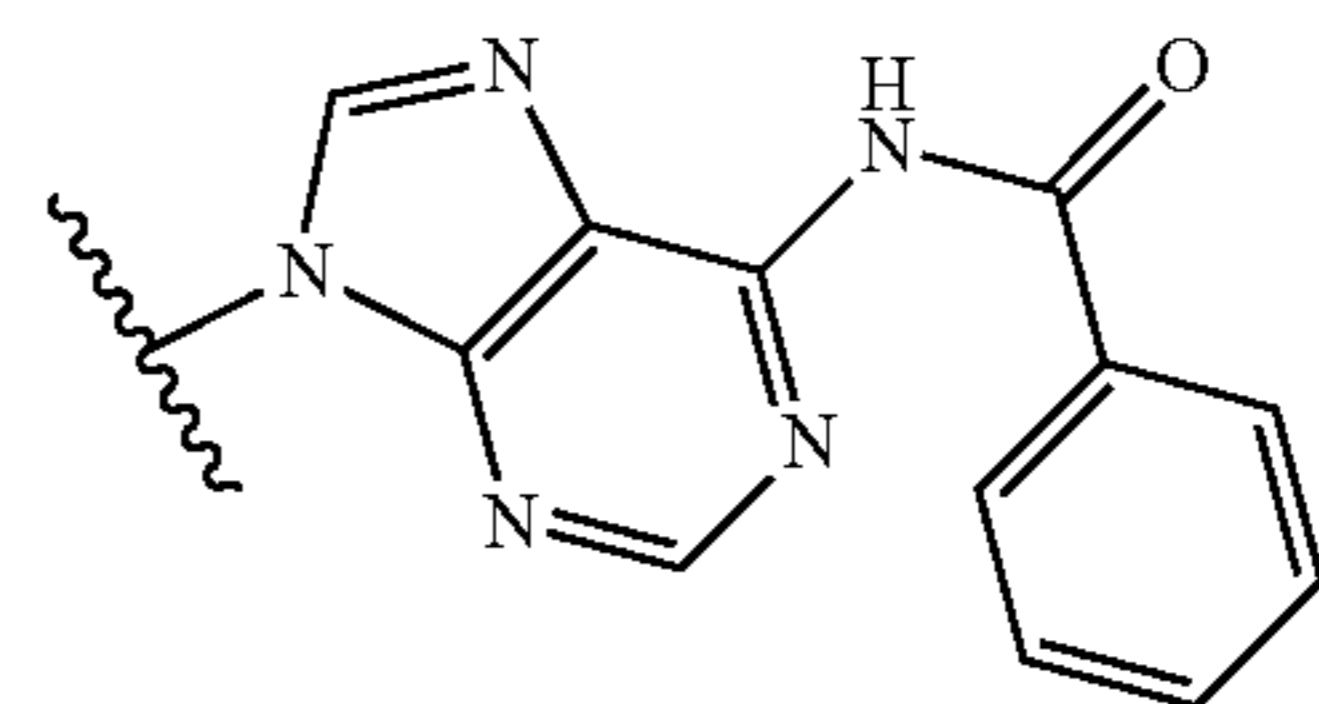
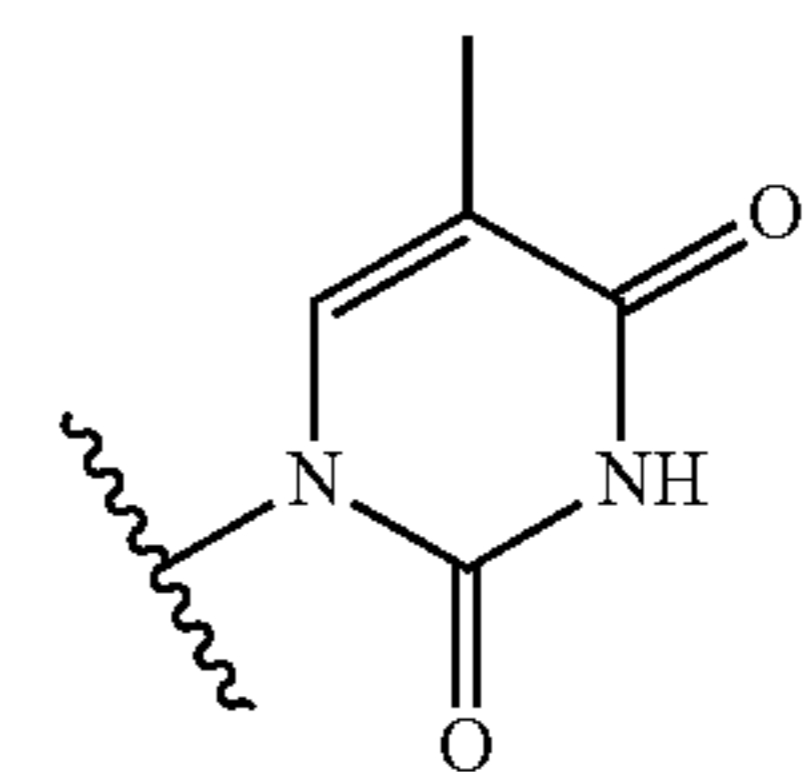
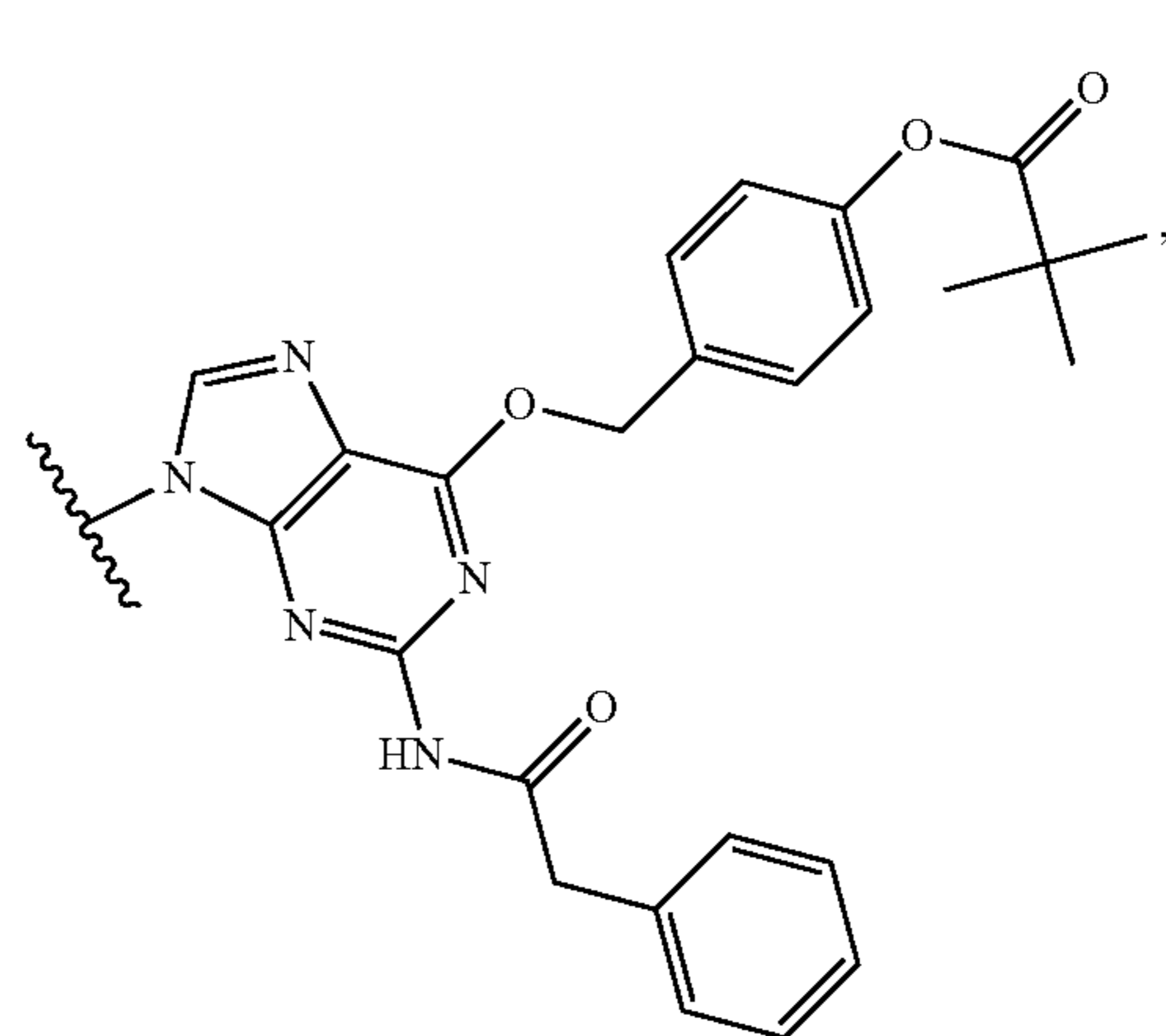
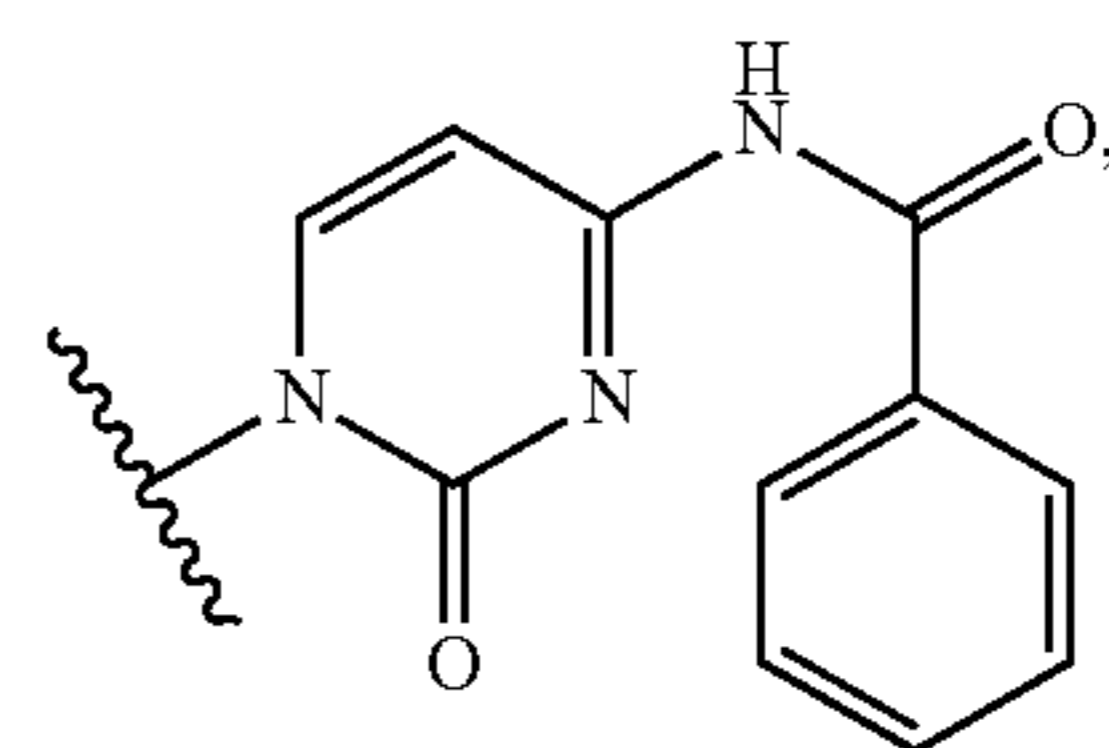


wherein R¹ is a support-medium, A is selected from the group consisting of:



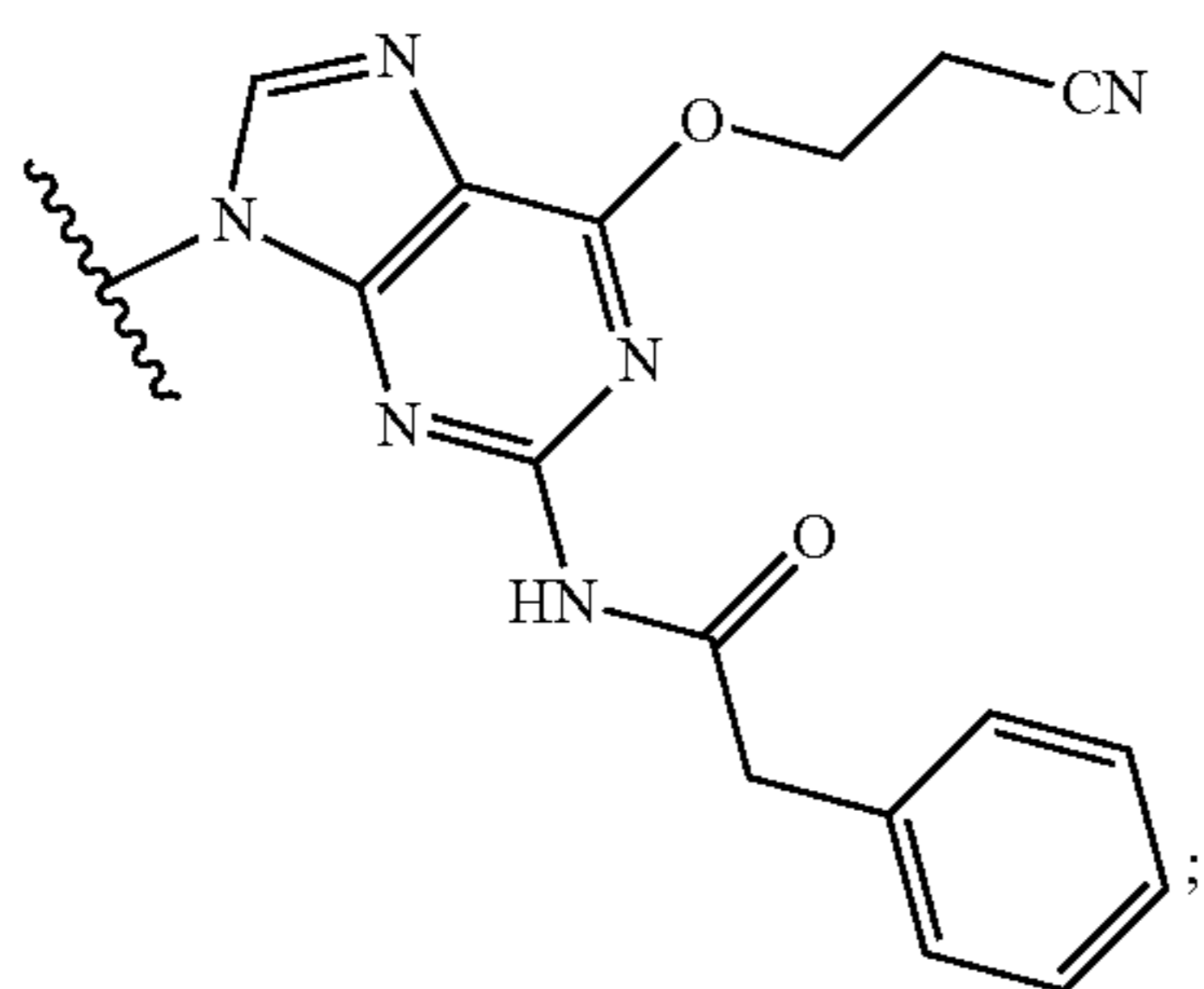
198

wherein R⁴ is selected from the group consisting of:



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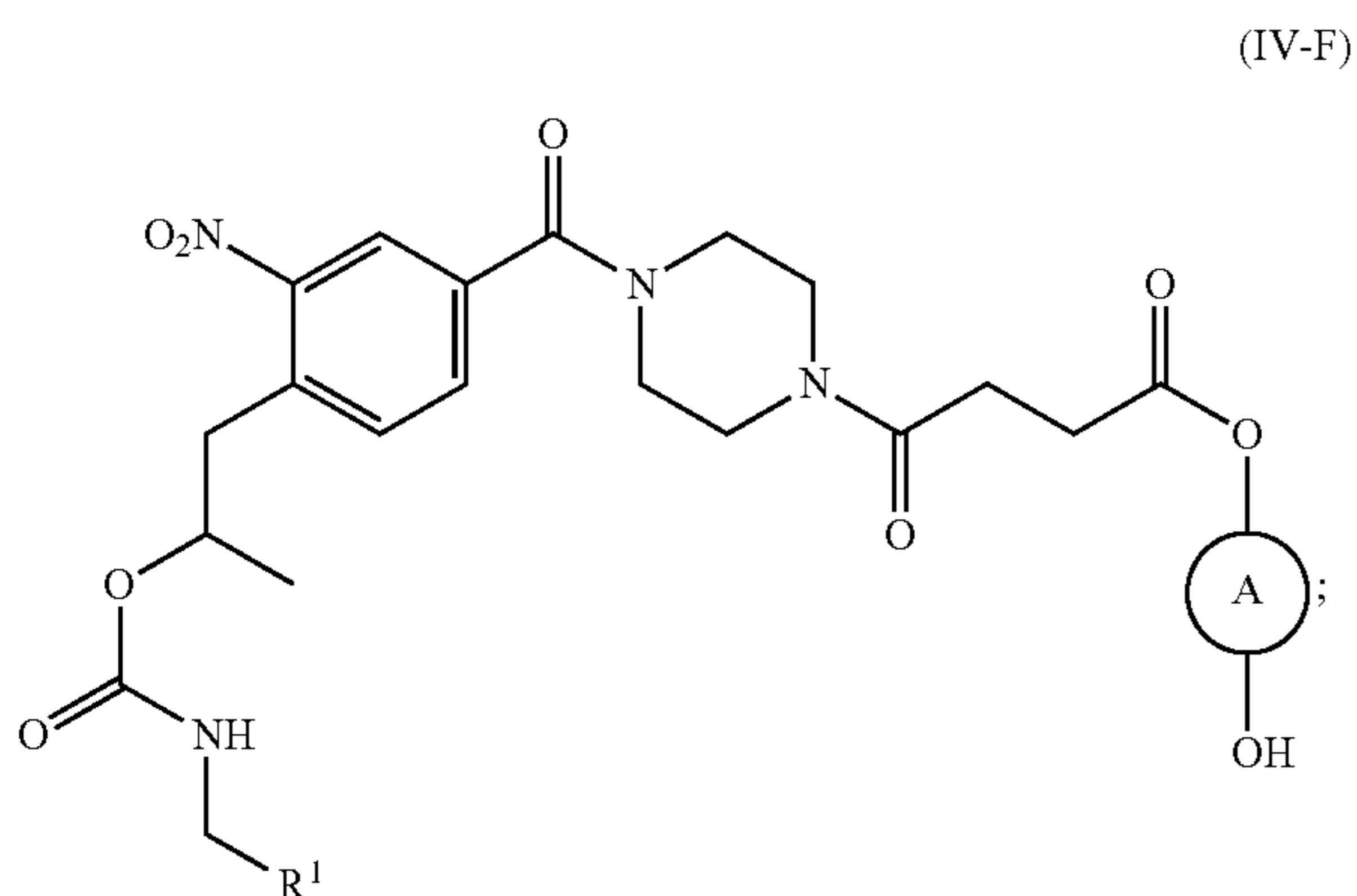
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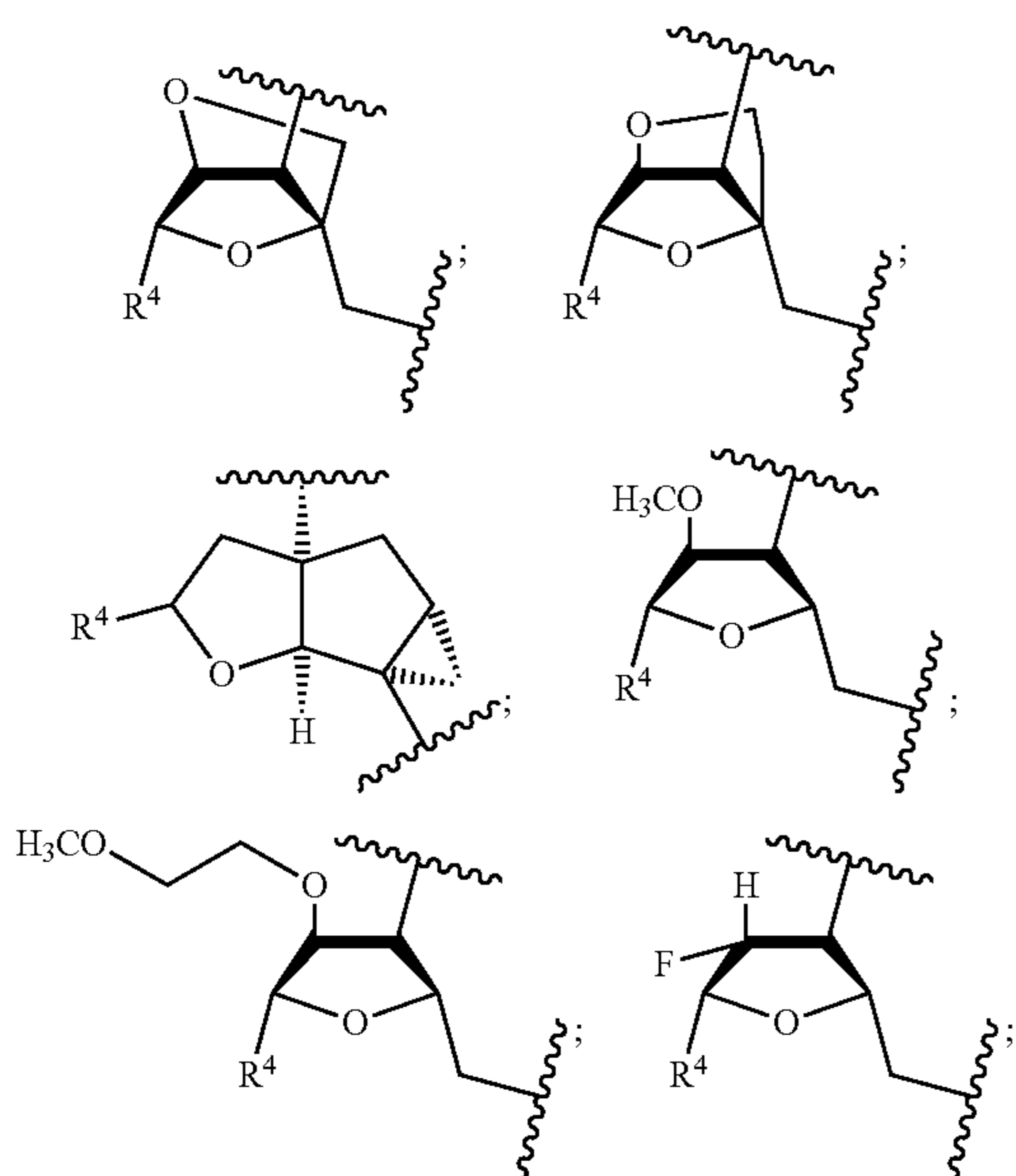
R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl

and trimethoxytrityl, and PG is a protecting group;

(b) contacting the compound of Formula (F3) with a deblocking agent to form a compound of Formula (IV-F):



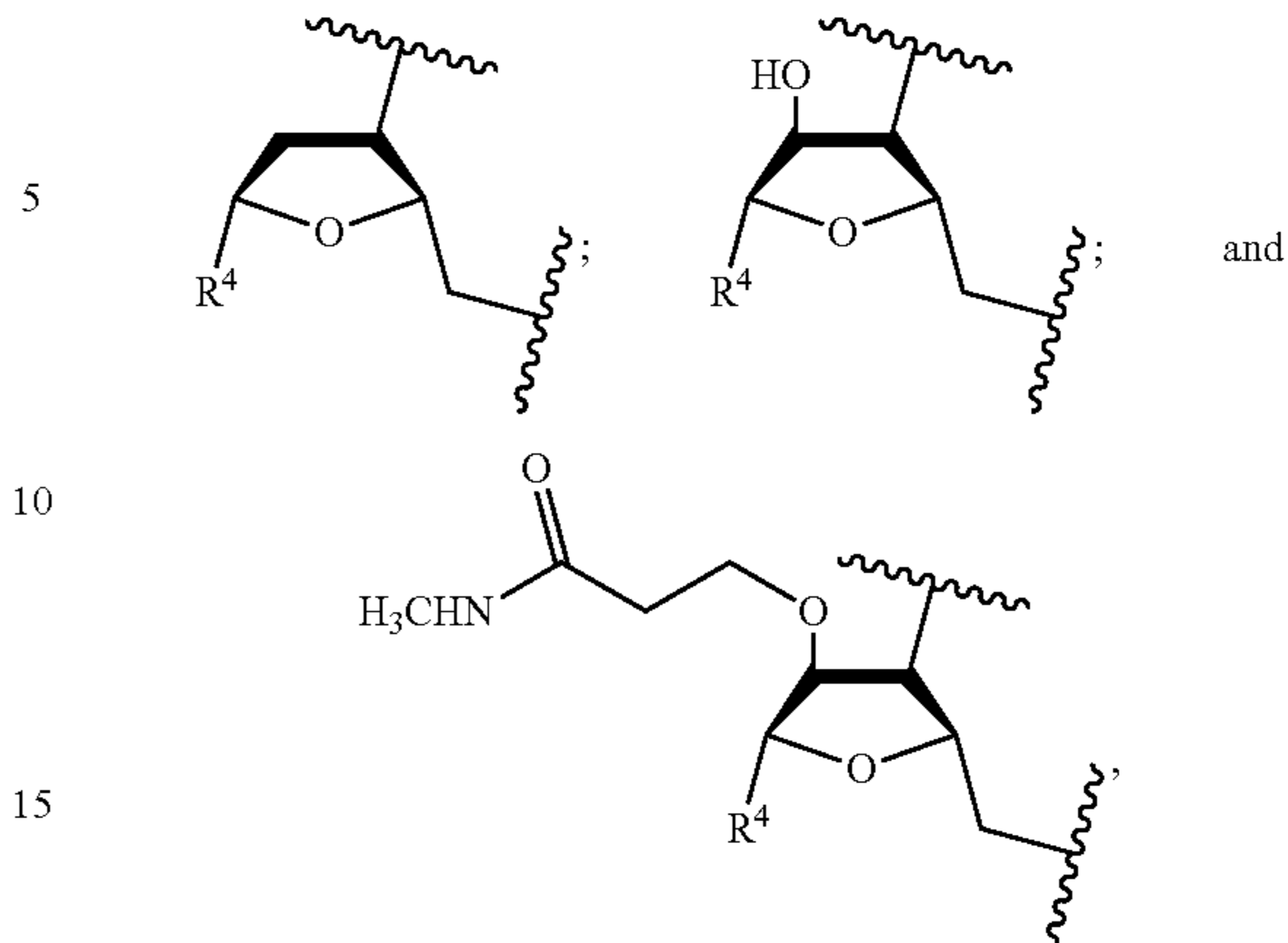
wherein R¹ is a support-medium, A is selected from the group consisting of:



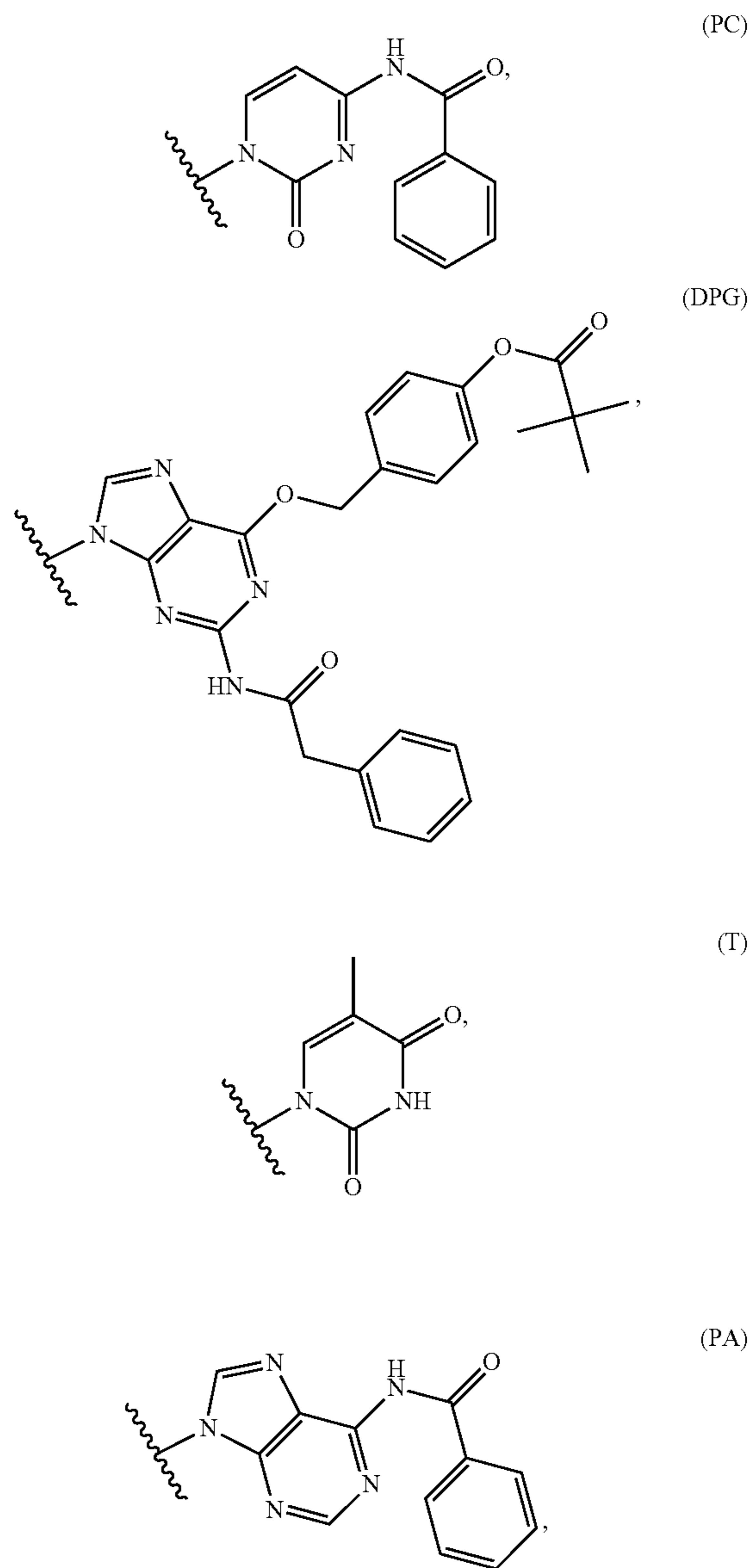
200

-continued

(PG)

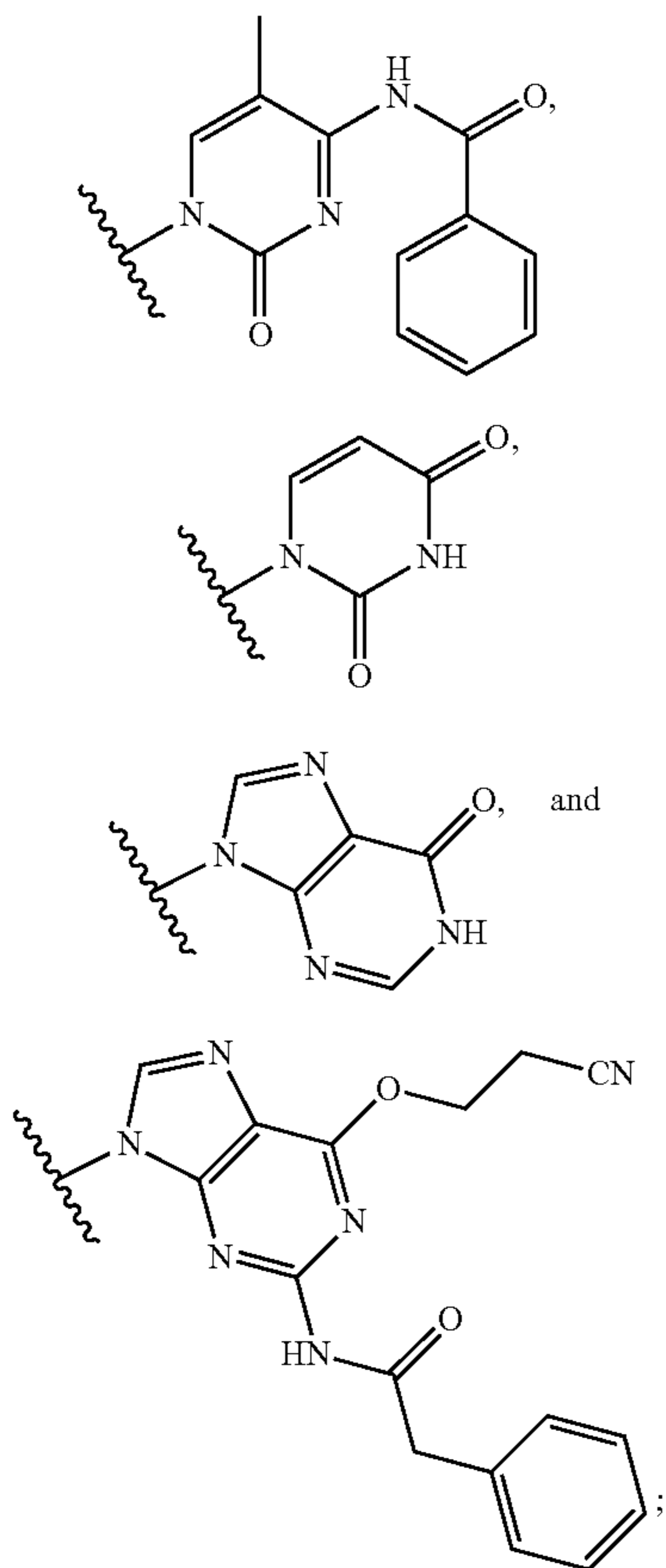


wherein R⁴ is selected from the group consisting of:

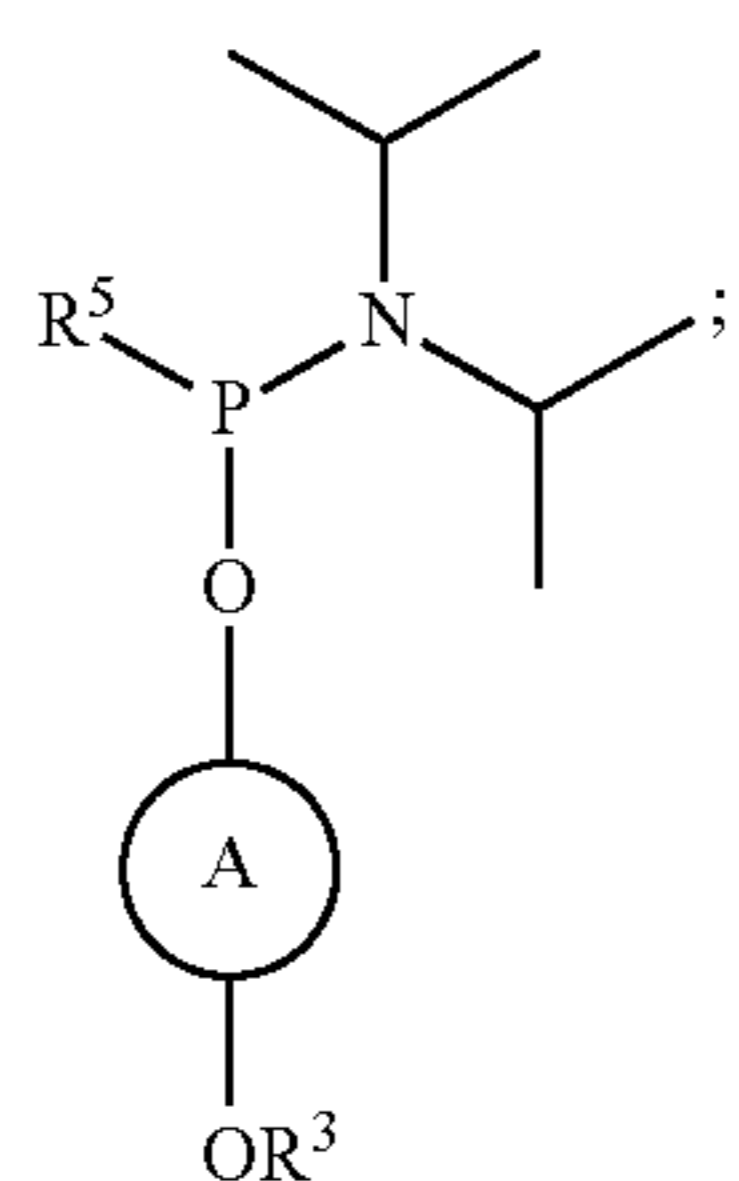


201

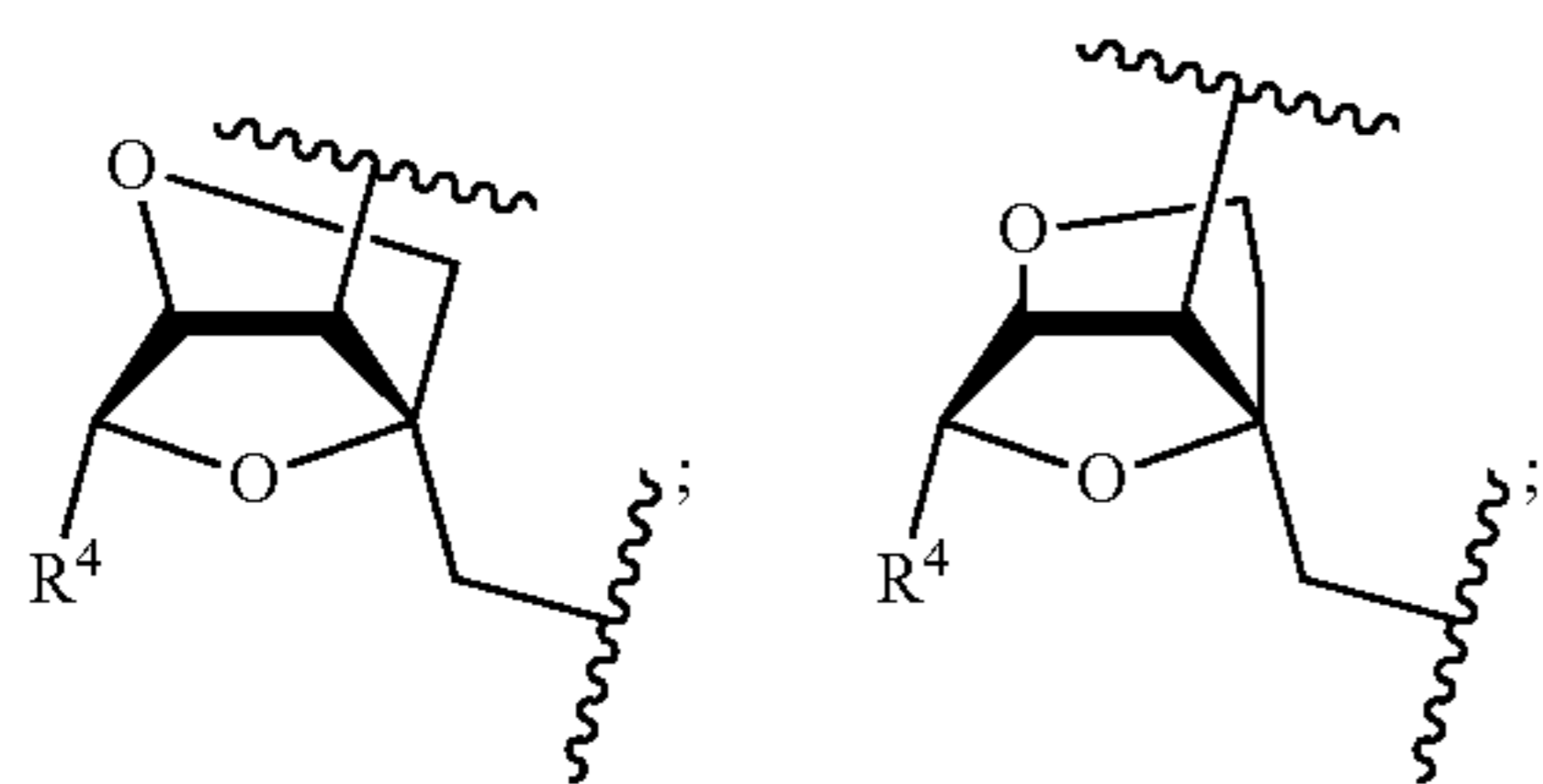
-continued



and PG is a protecting group;
(c) contacting the compound of Formula (IV-F) with a compound of Formula (F4):

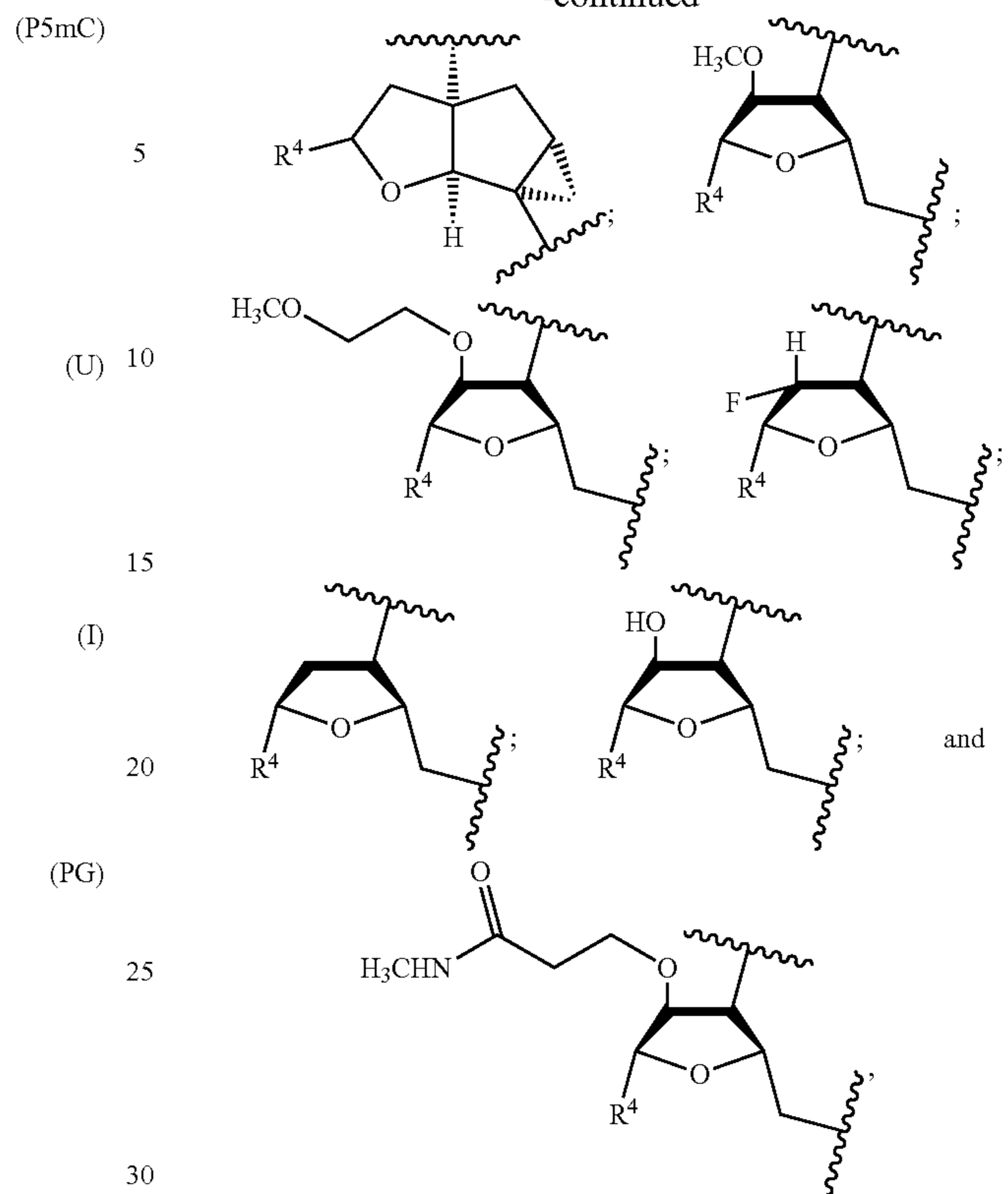


wherein R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN, A is selected from the group consisting of:

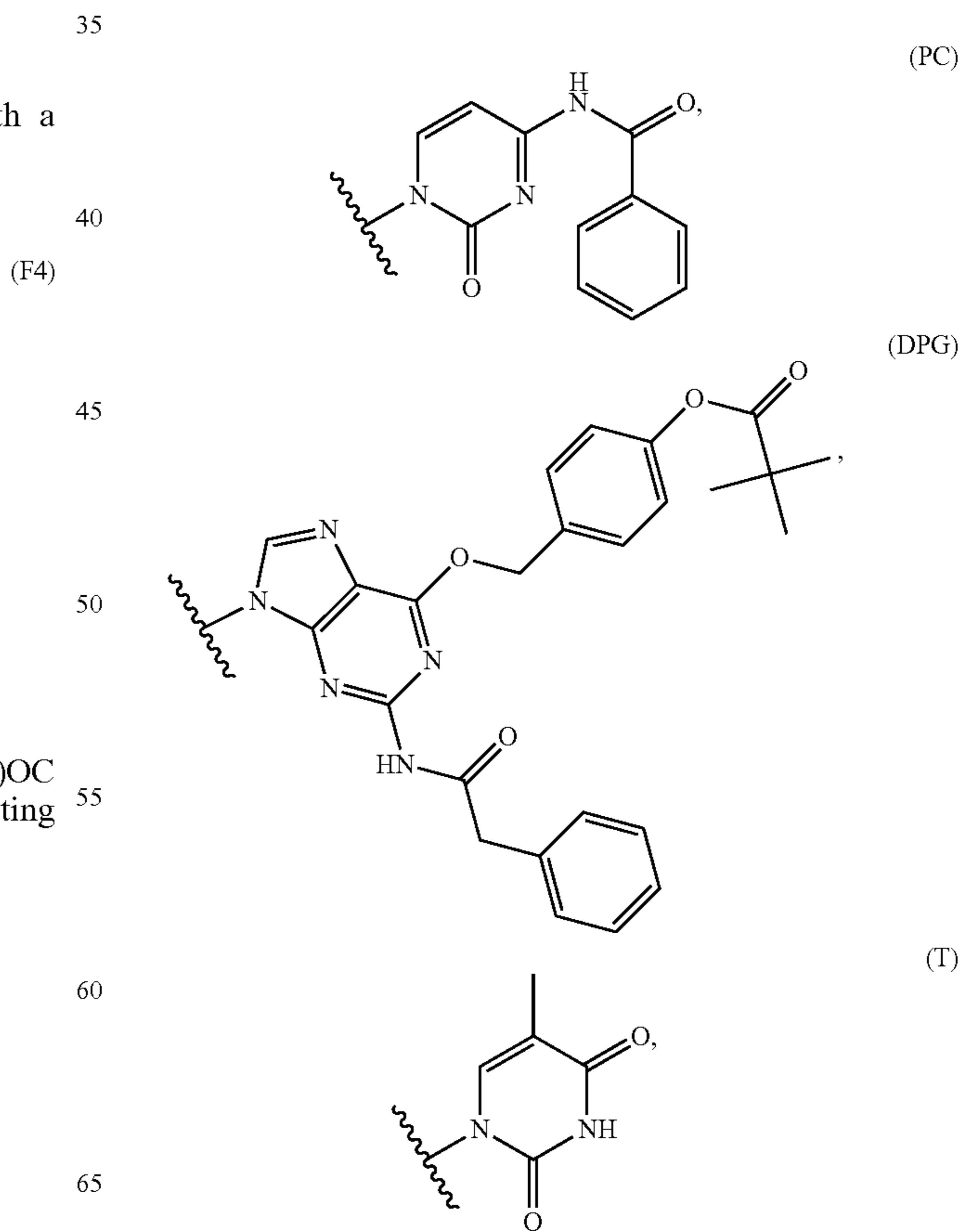


202

-continued

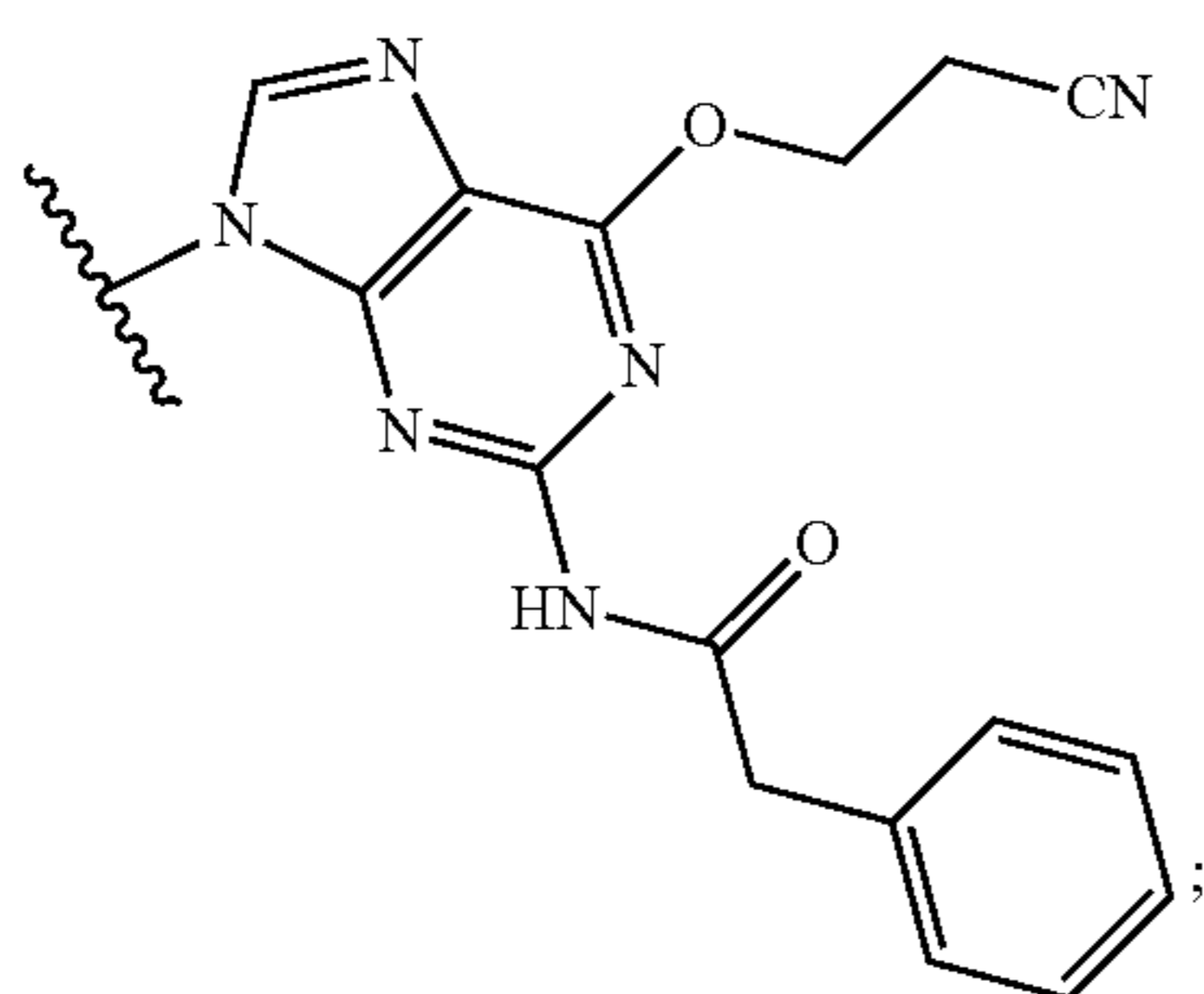
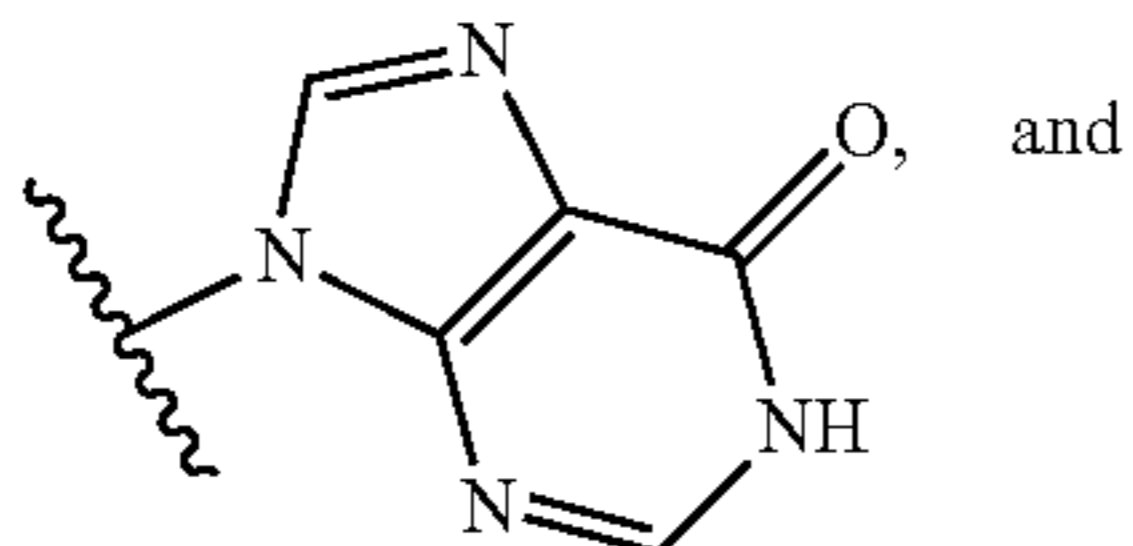
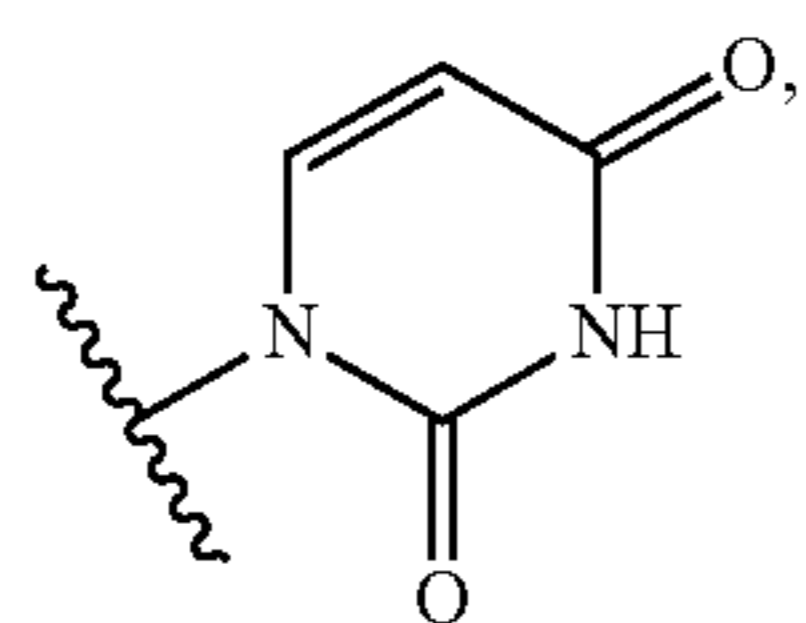
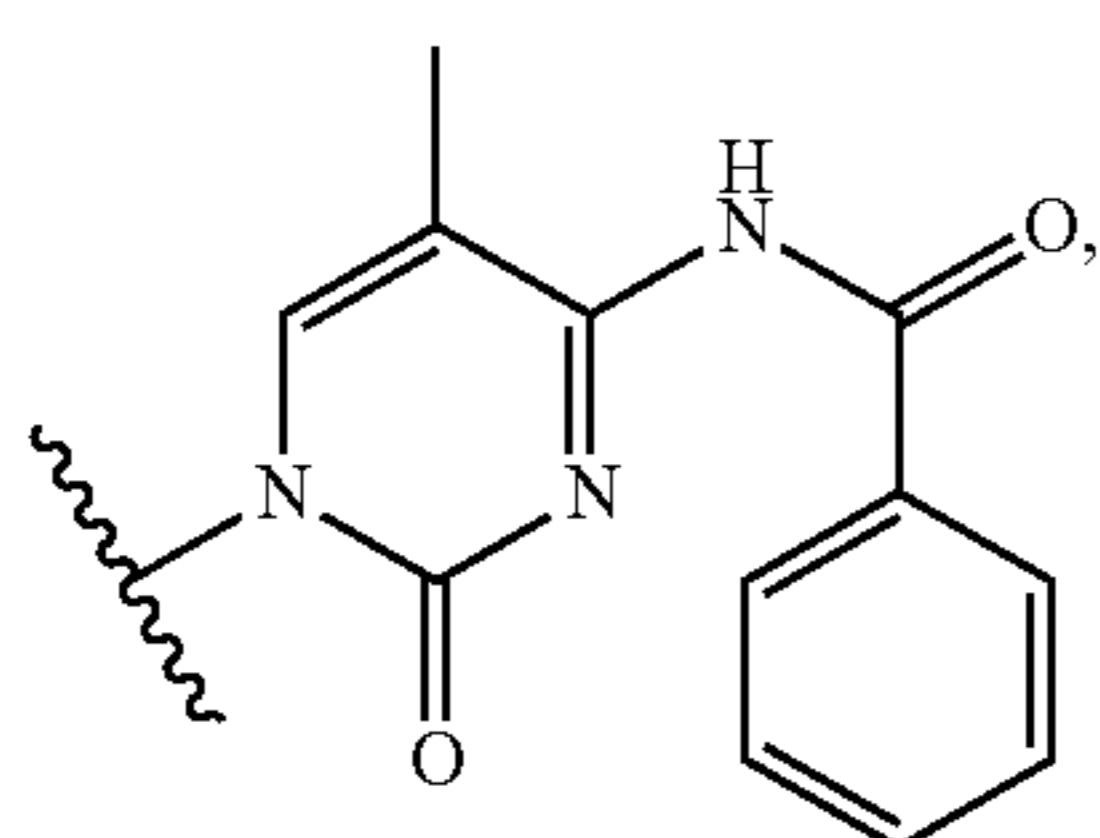
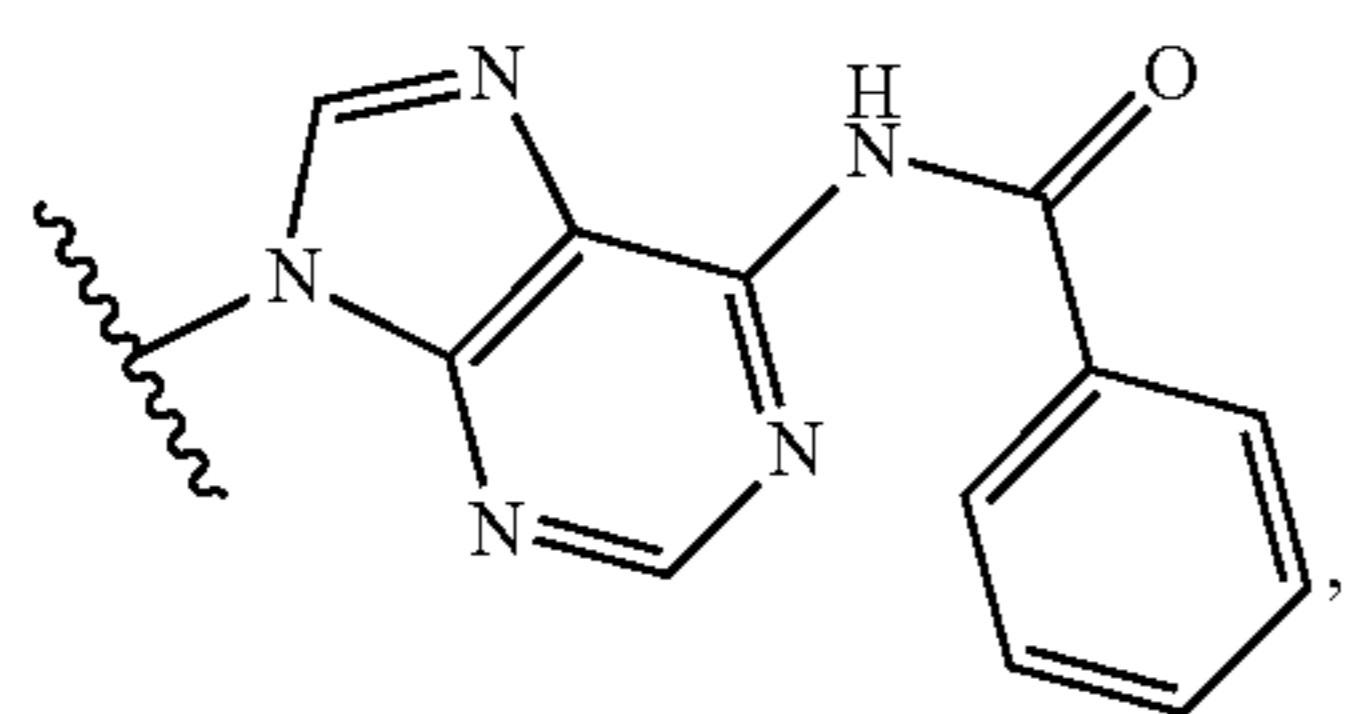


wherein R⁴ is selected from the group consisting of:



203

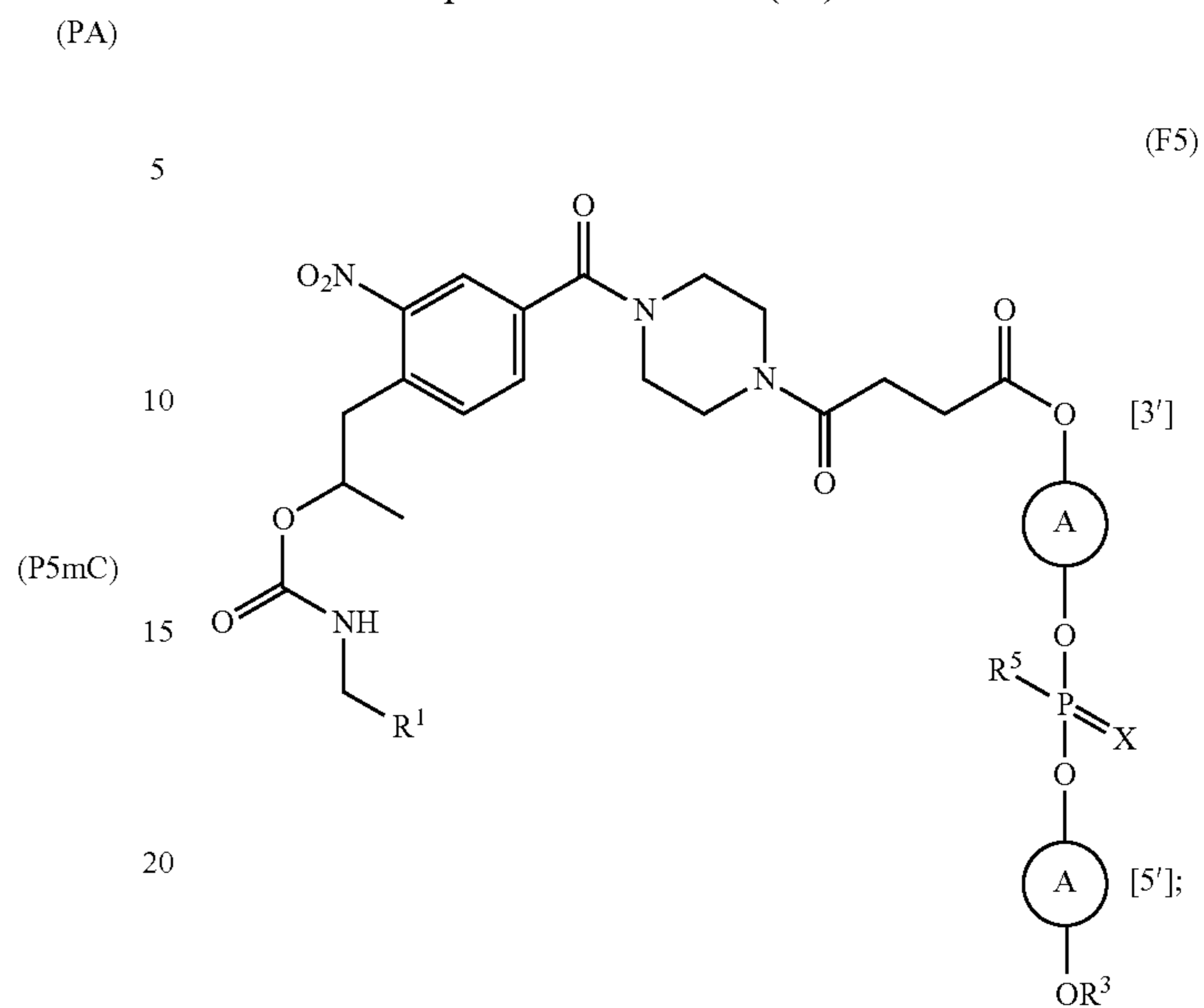
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R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and PG is a protecting group,

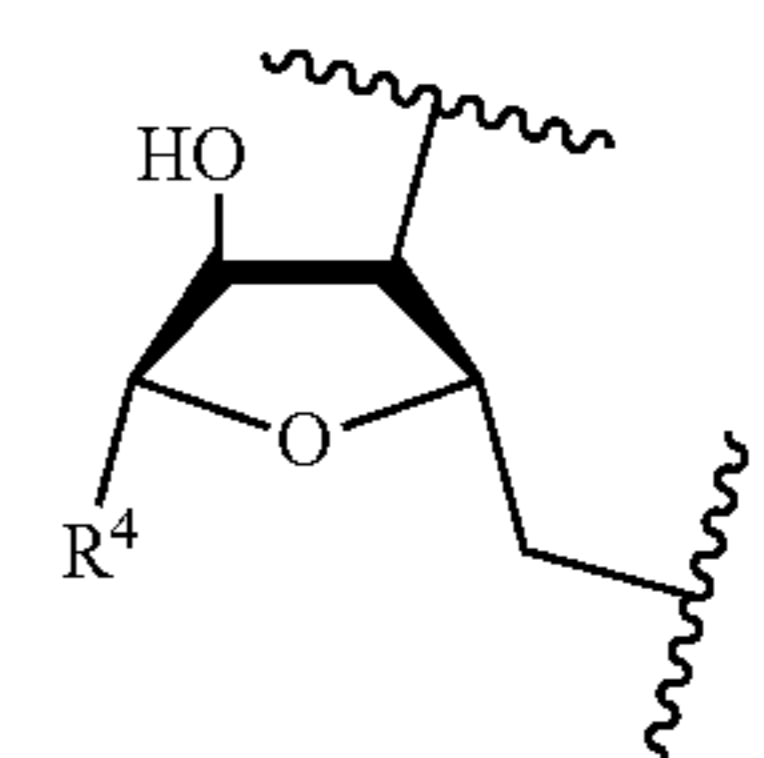
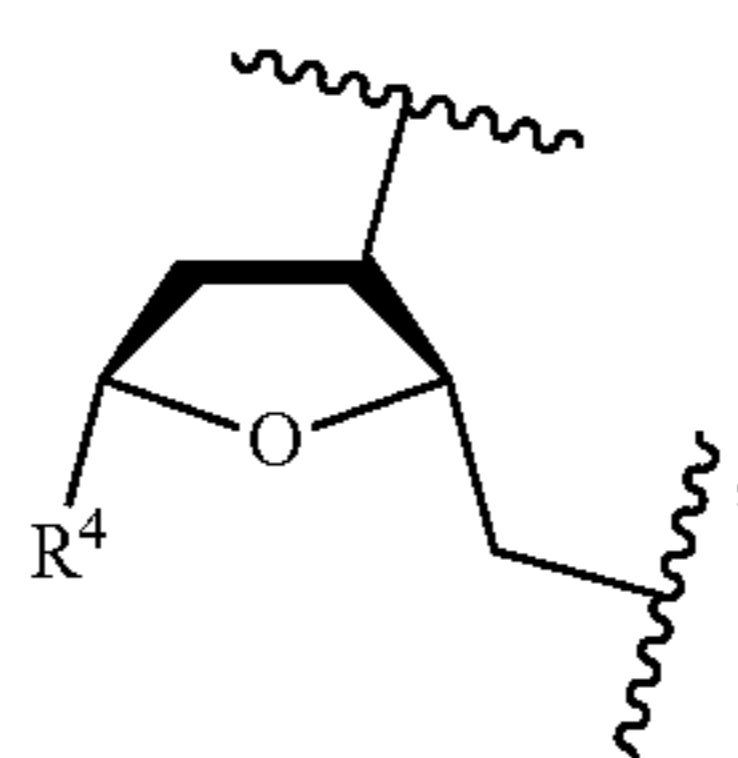
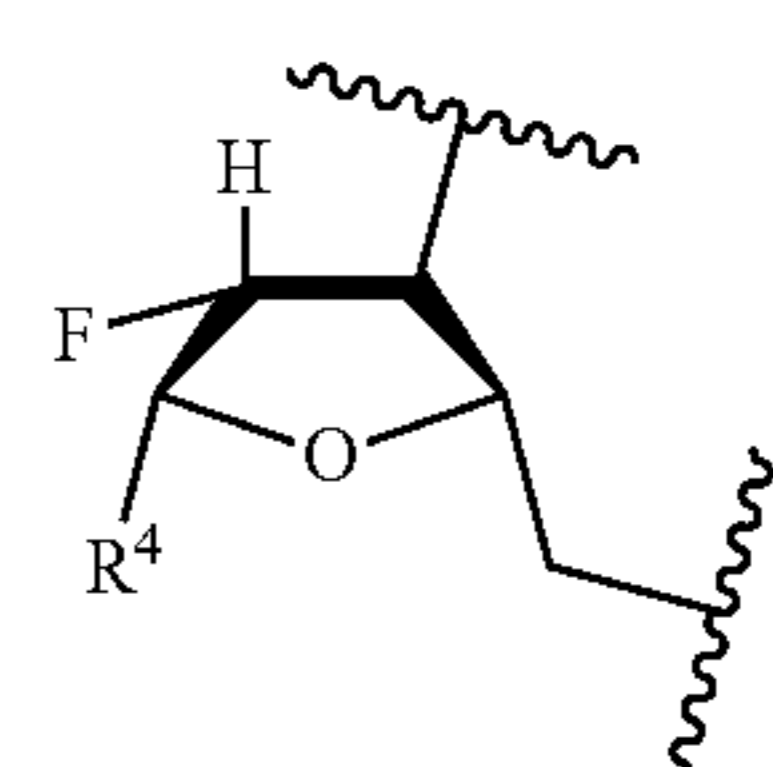
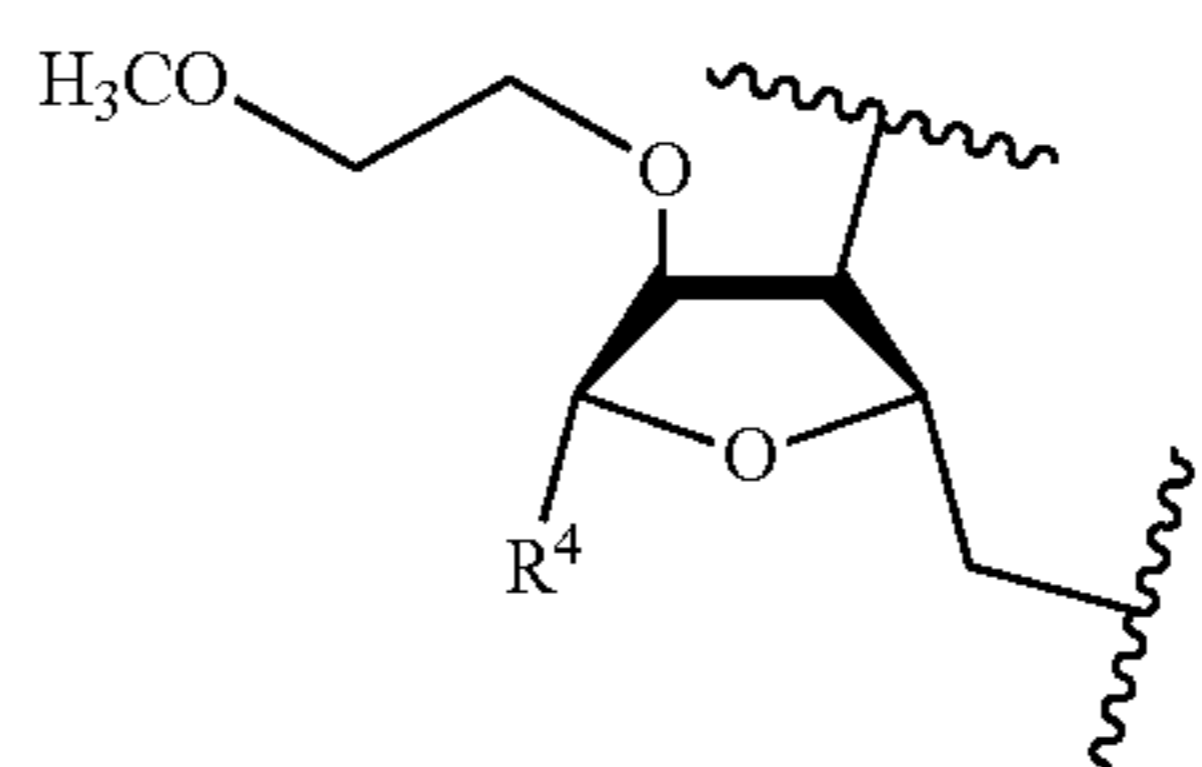
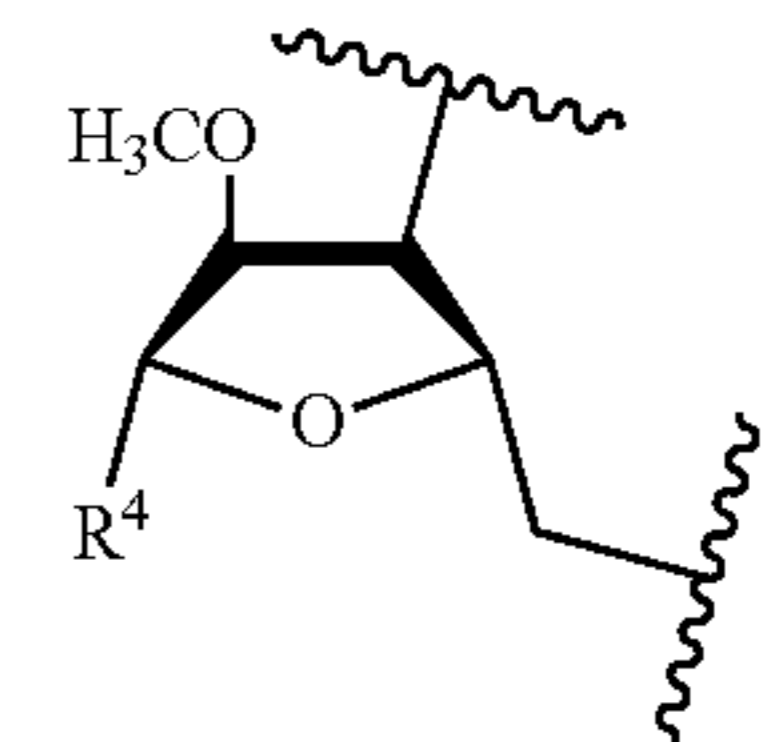
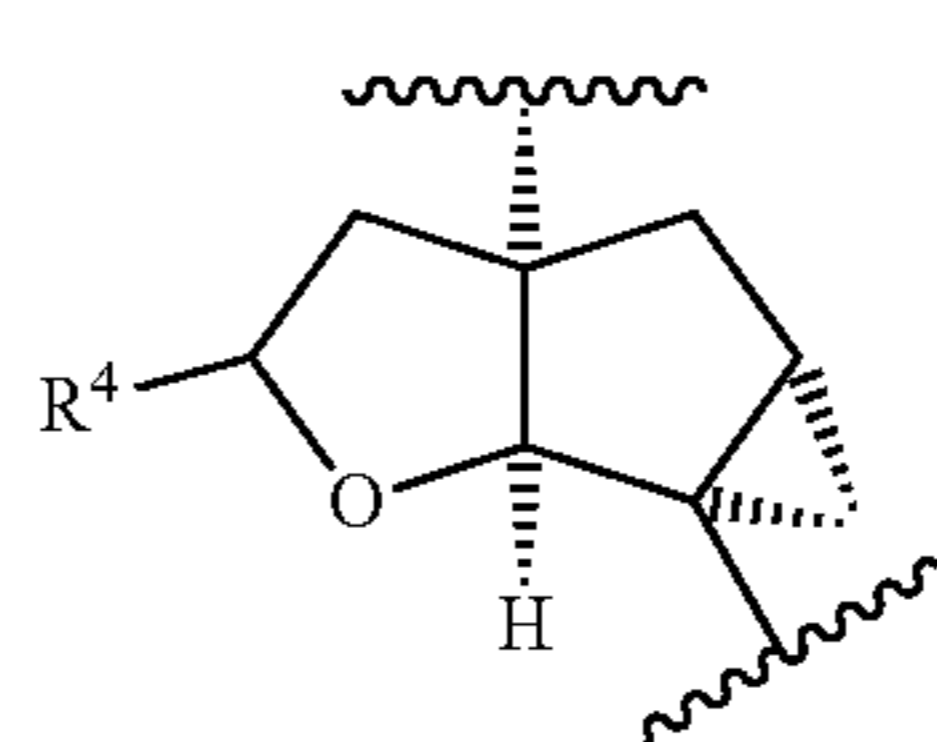
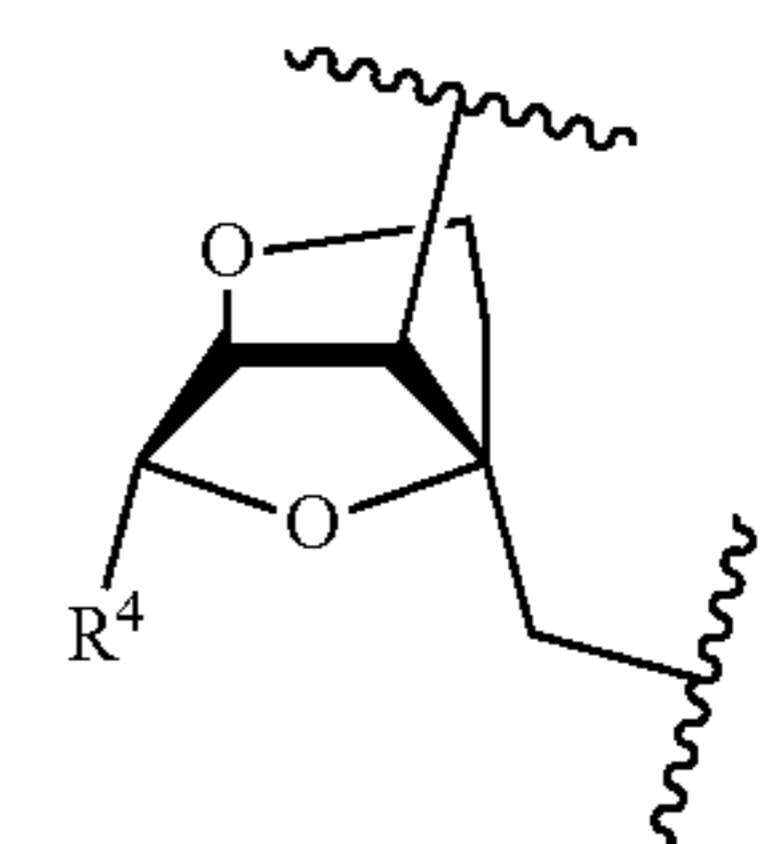
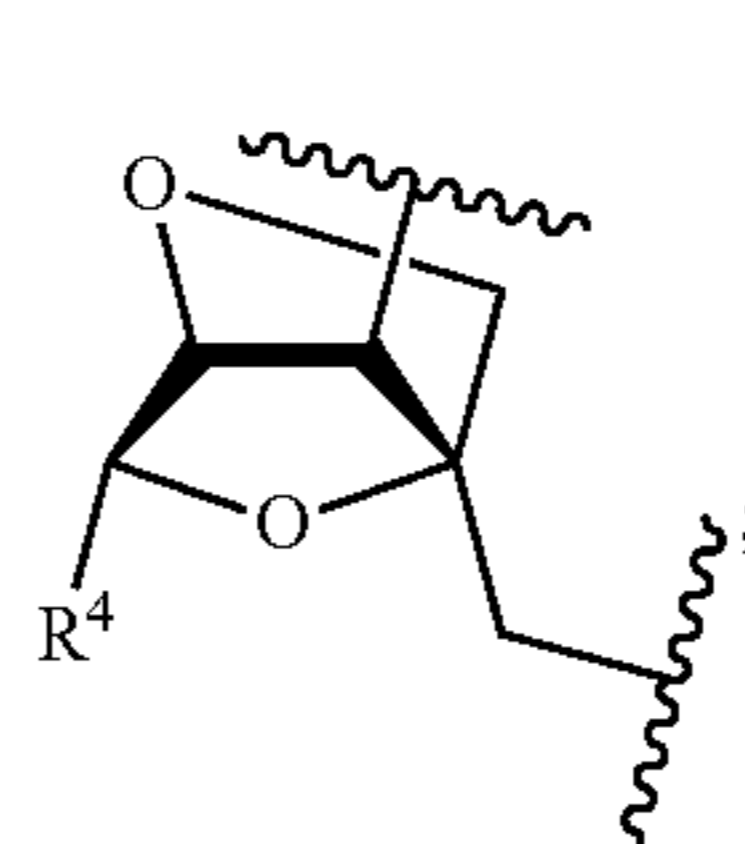
204

to form a compound of Formula (F5):



wherein R¹ is a support-medium,

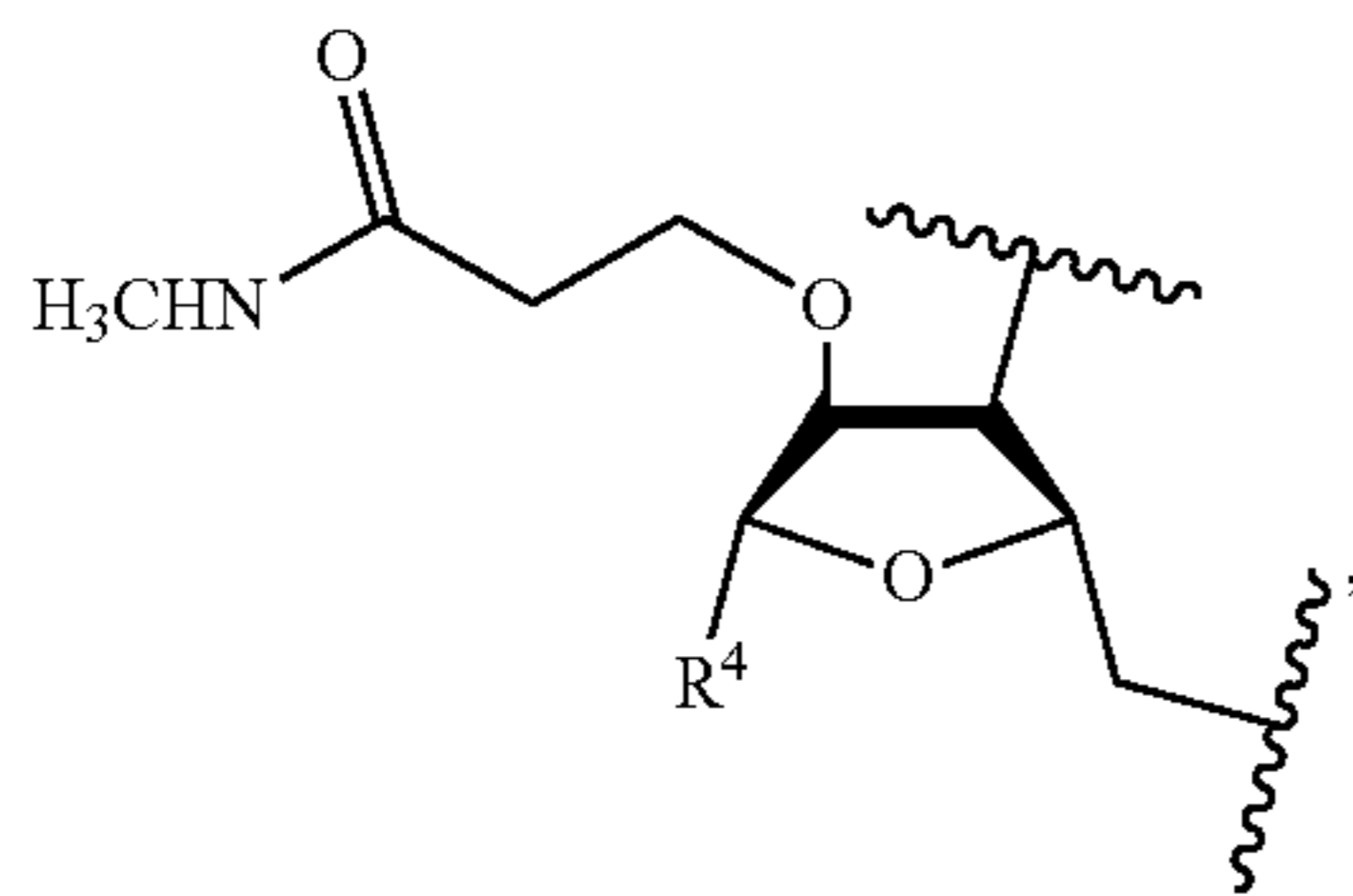
each A is, independently at each occurrence, selected from the group consisting of:



and

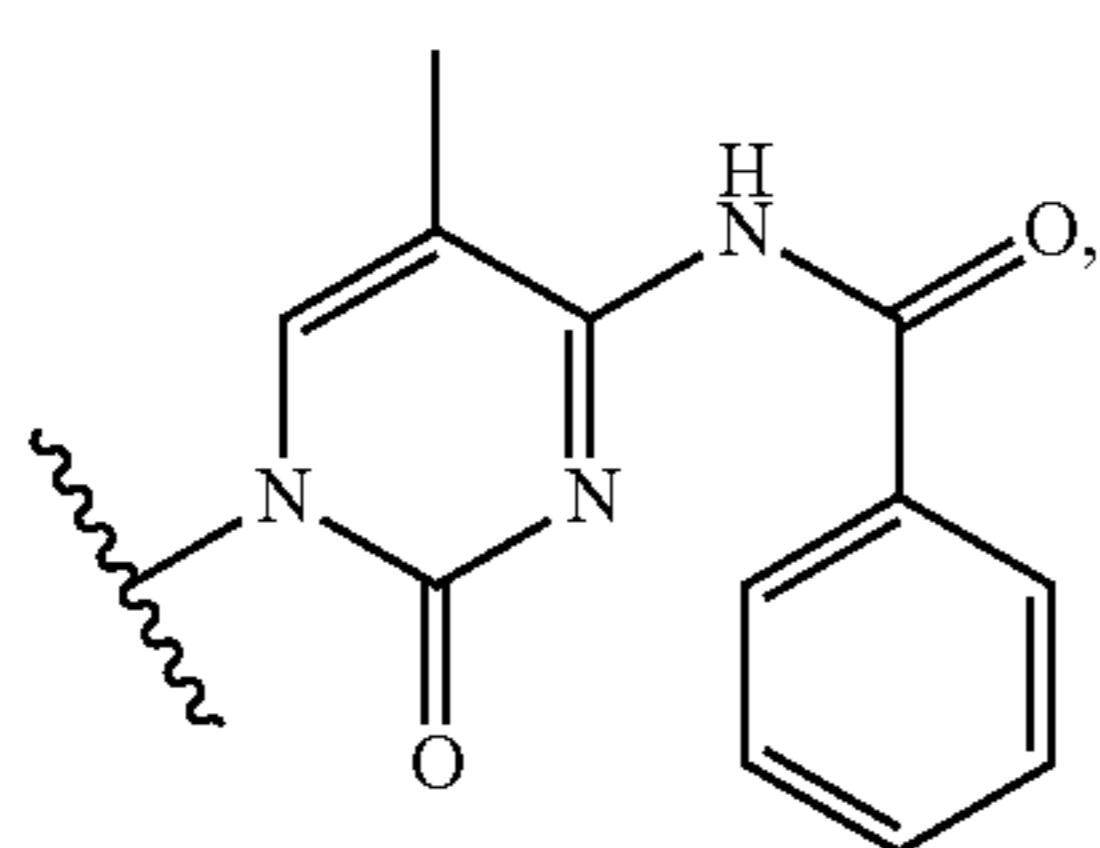
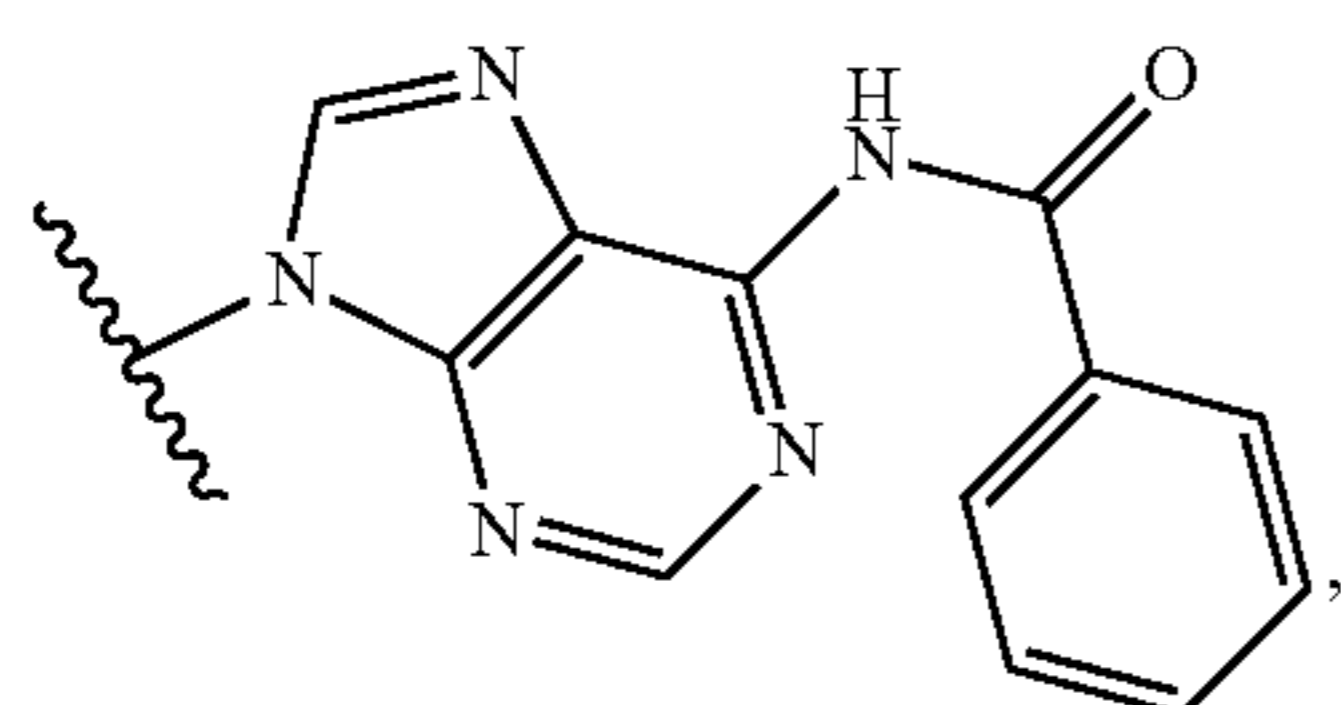
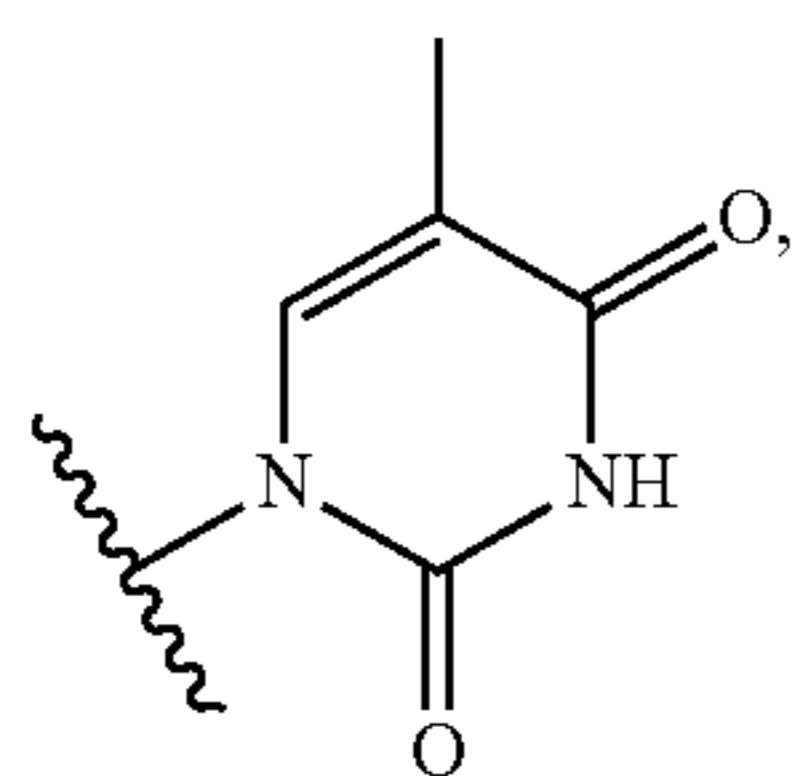
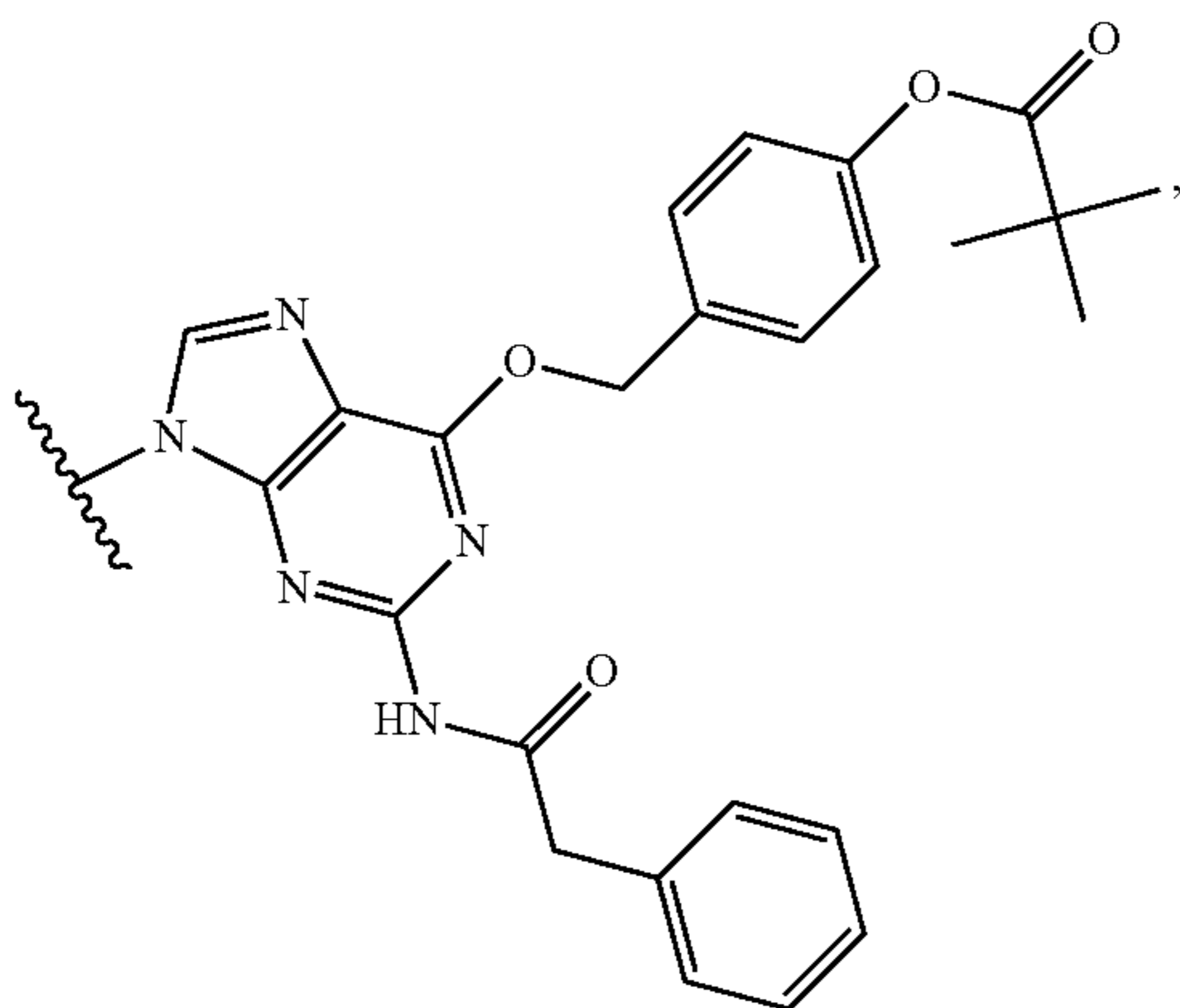
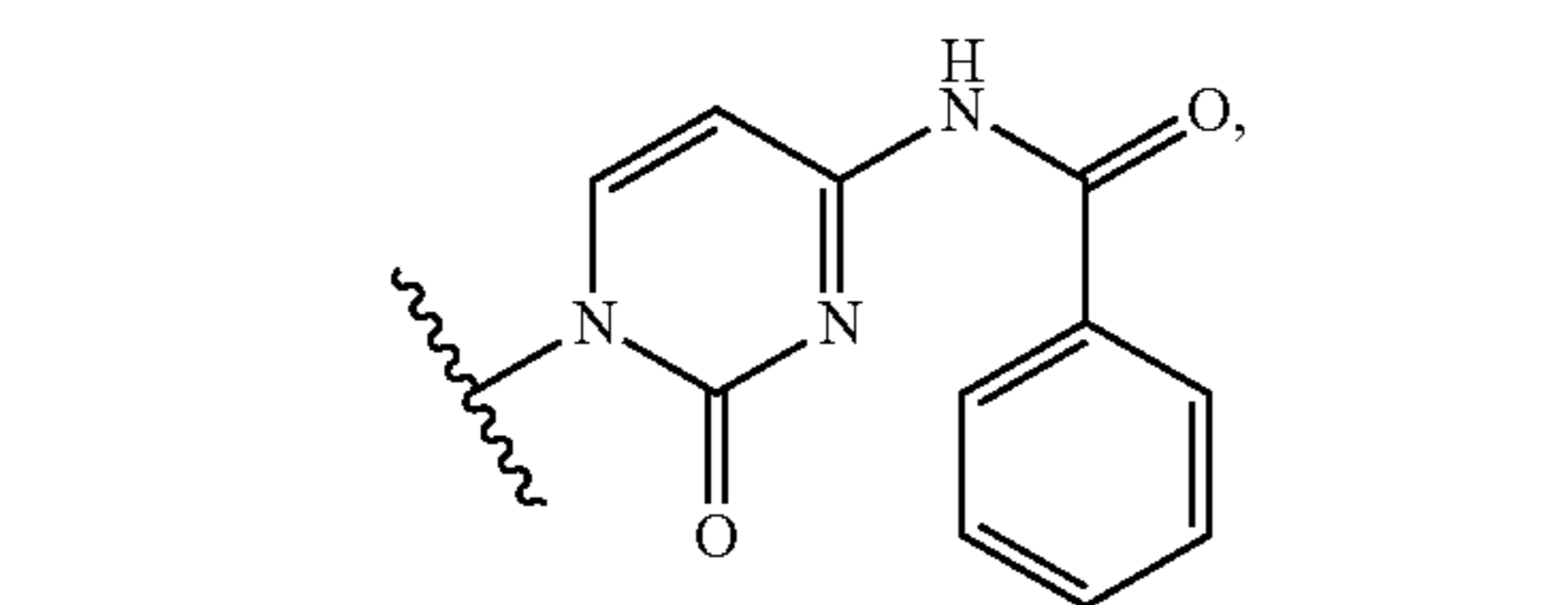
205

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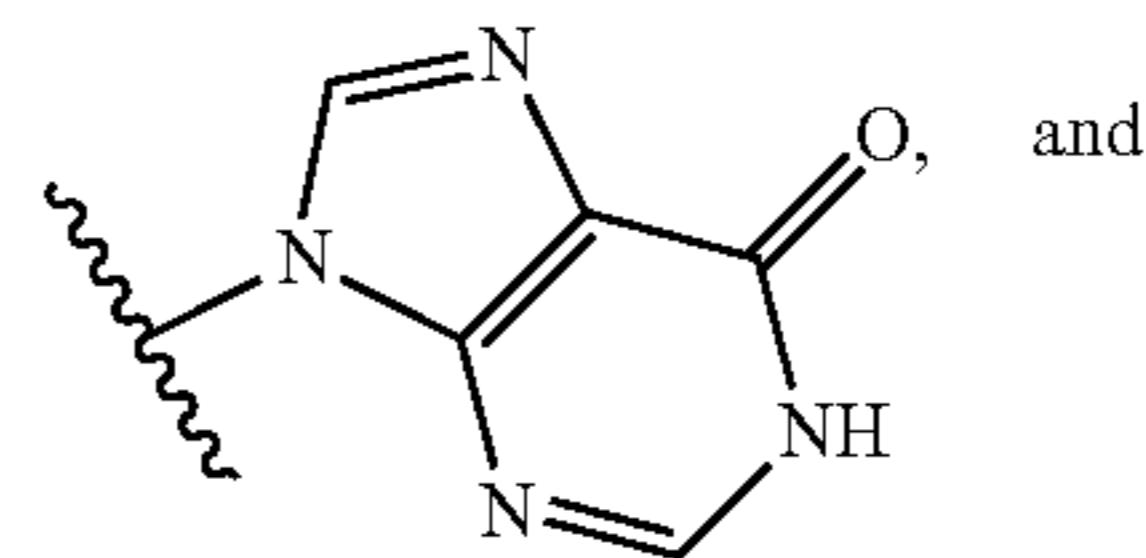
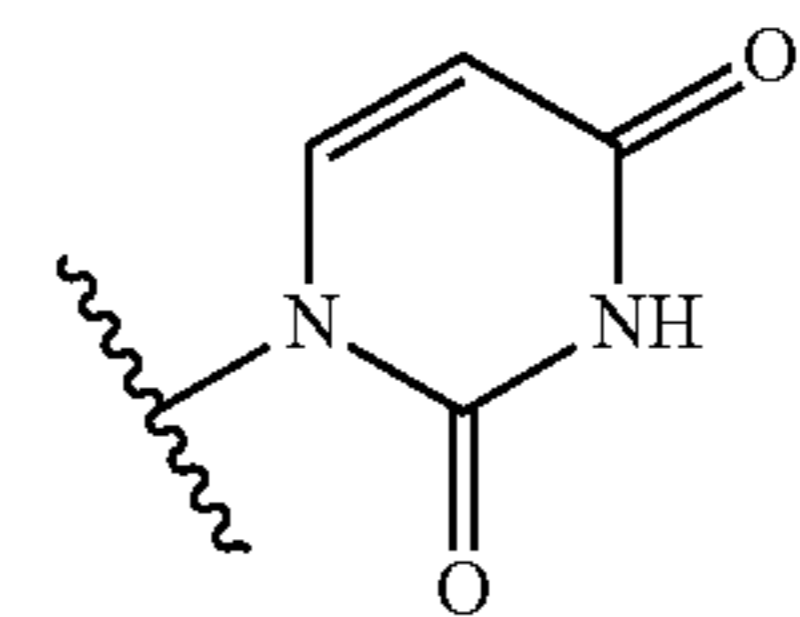
and

each R⁴ is, independently at each occurrence, selected from the group consisting of:



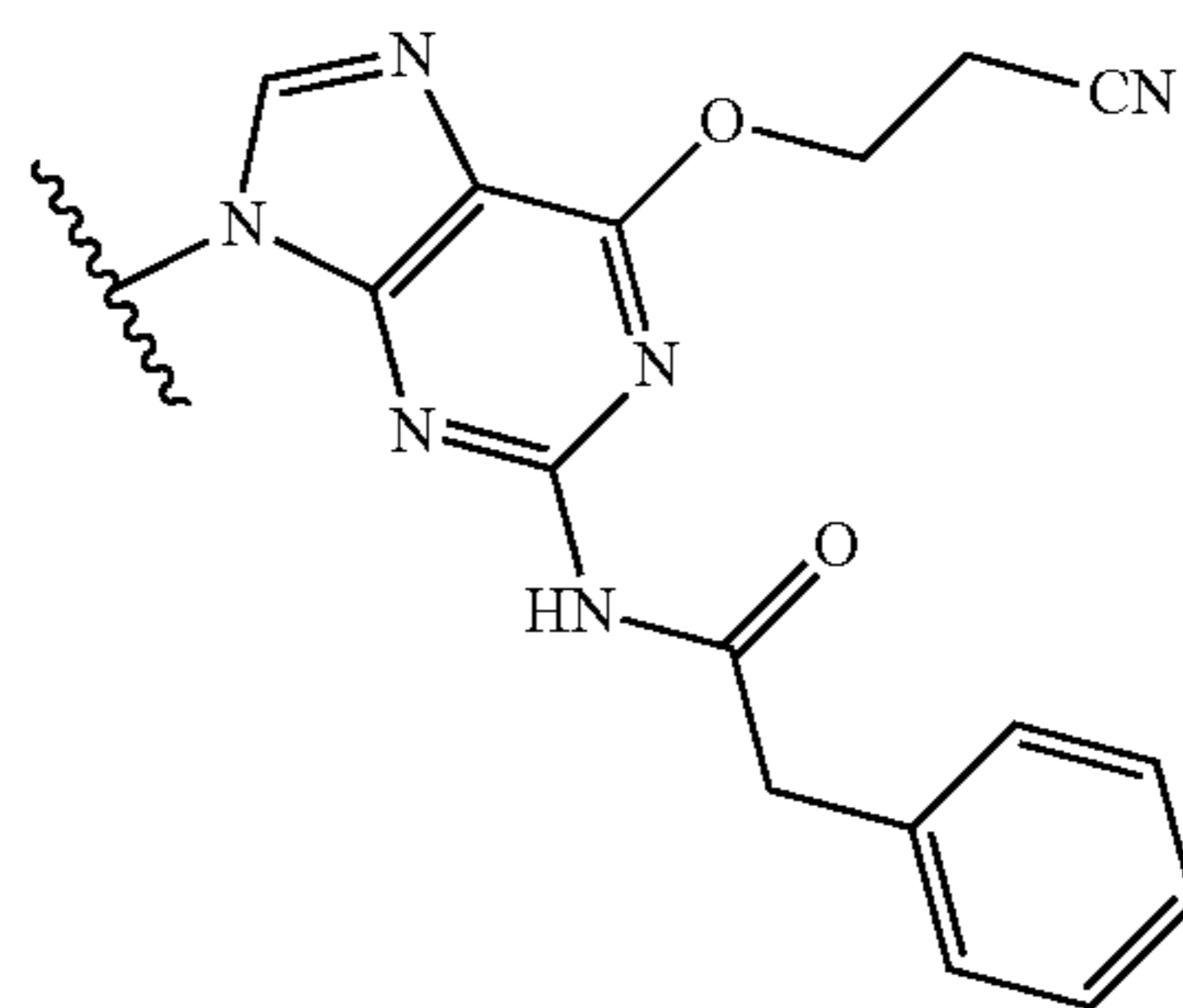
206

-continued



(PC)

20



25

(DPG)

30

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl,

35

X is O or S, and

R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN,

40

wherein where X is S, then step (d) further comprises a sulfurization step, or where X is O, then step (d) further comprises an oxidation step;

(T)

(e) performing n-2 iterations of the sequential steps of:

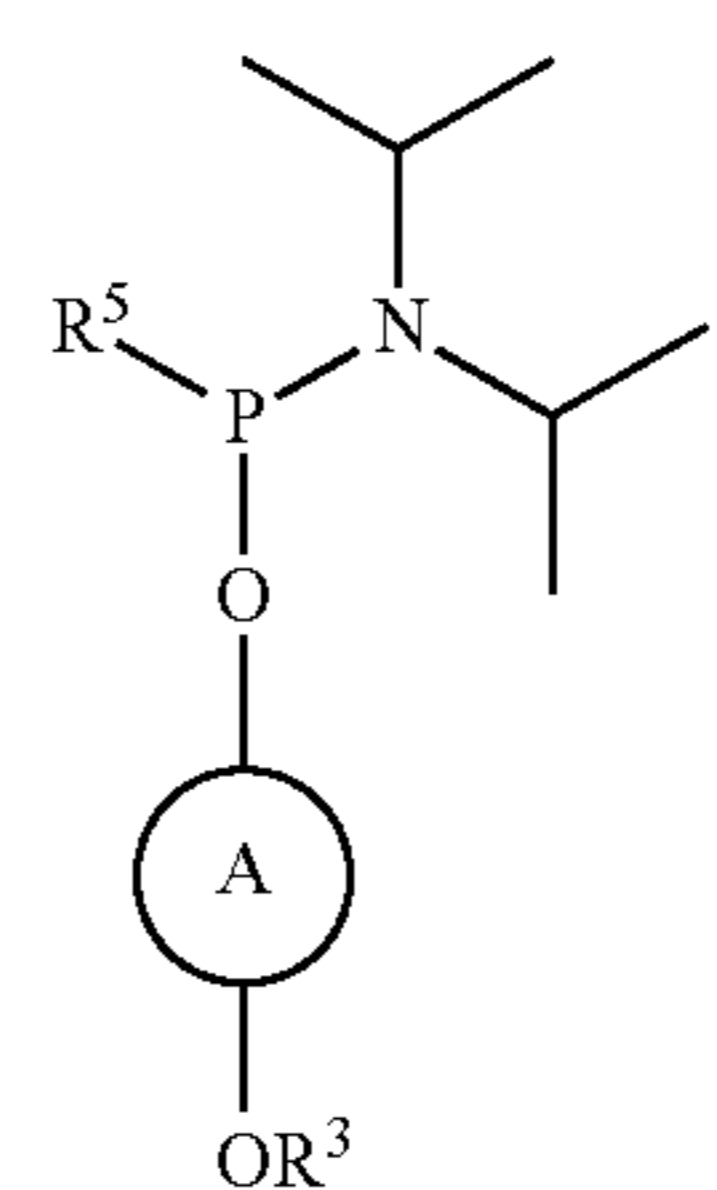
45

(e1) contacting the product formed by the immediately prior step with a deblocking agent; and

(e2) contacting the compound formed by the immediately prior step with a compound of Formula (F8):

(PA)

50



55

(P5mC)

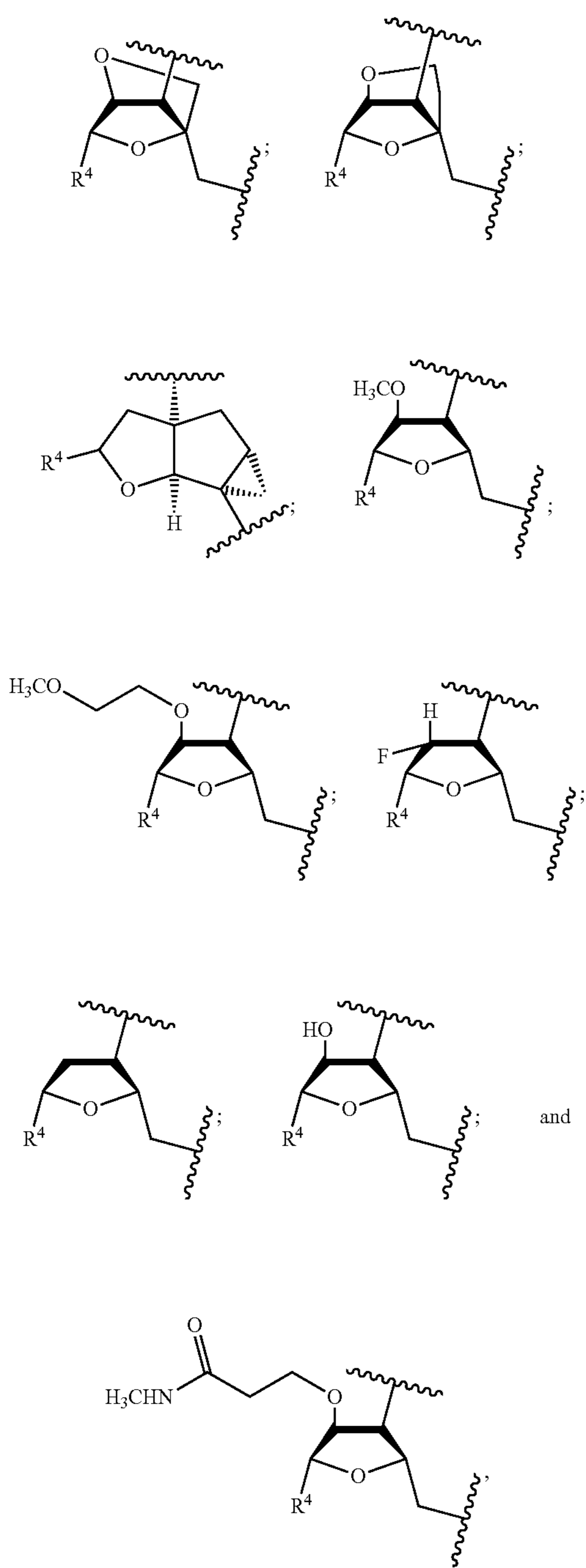
60

wherein R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN,

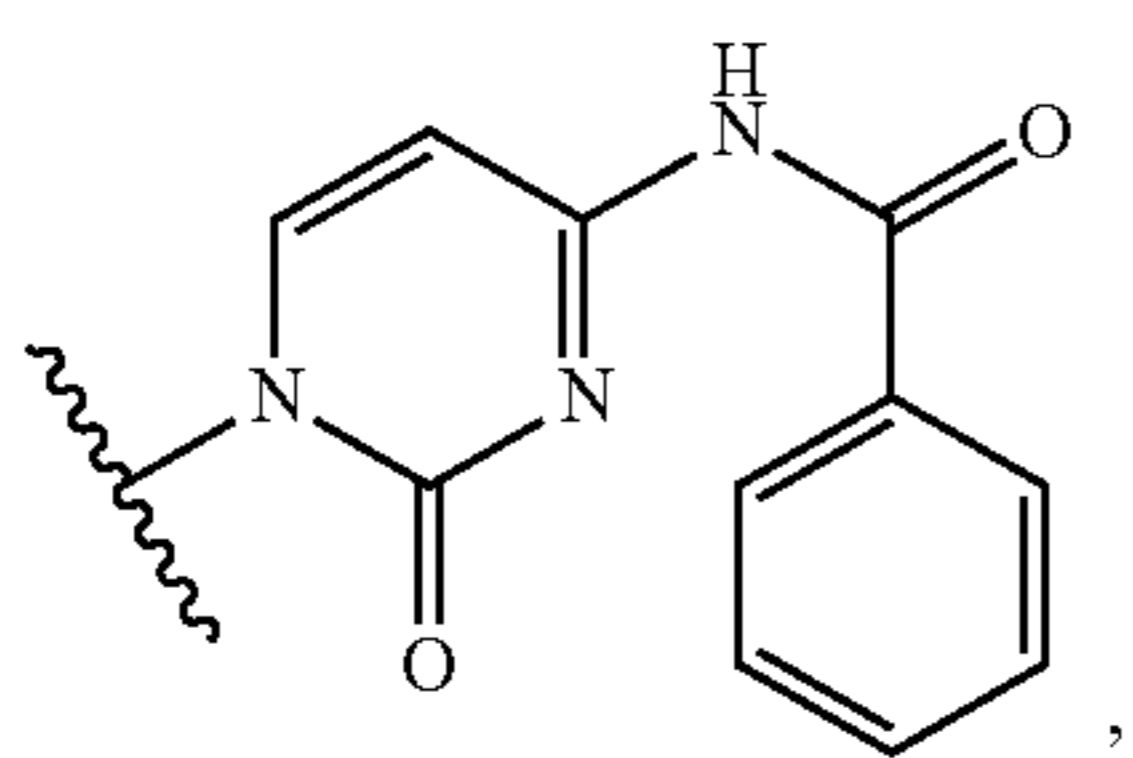
65

each A is, independently for each iteration of (F8), selected from the group consisting of:

207

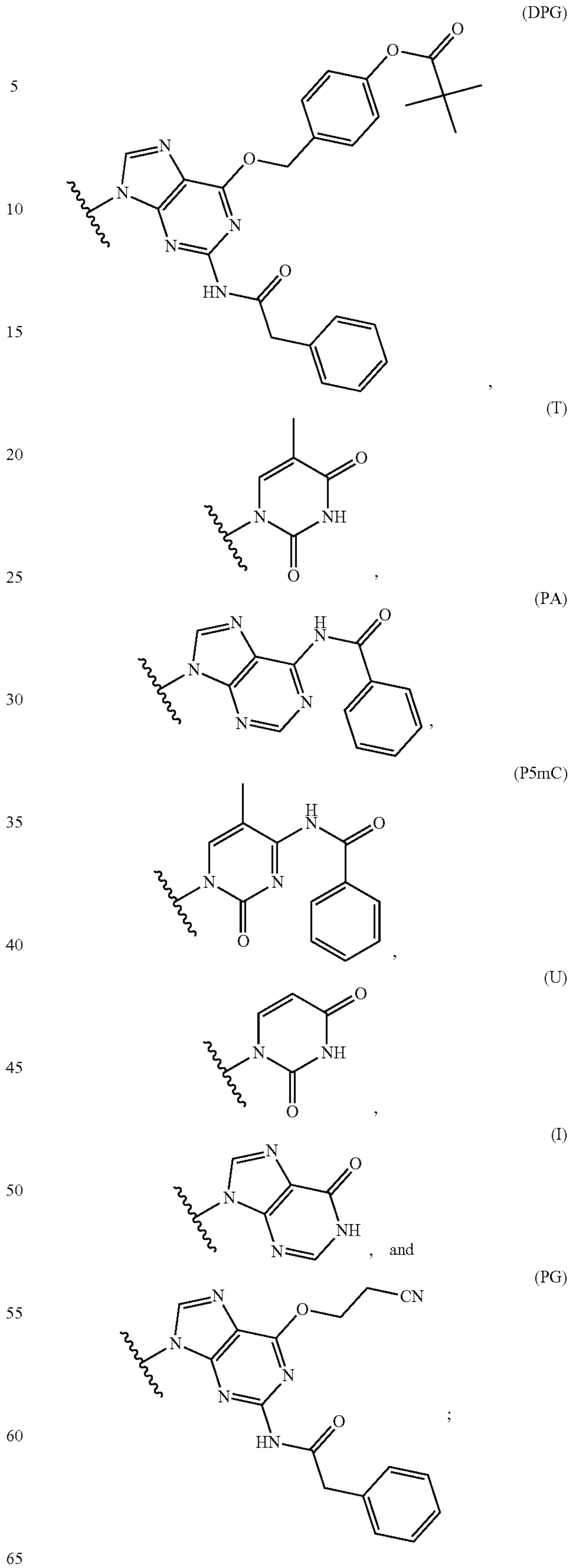


wherein R⁴ is, independently for each iteration of (F8), selected from the group consisting of:



208

-continued



(DPG)

(T)

(PA)

(P5mC)

(U)

(I)

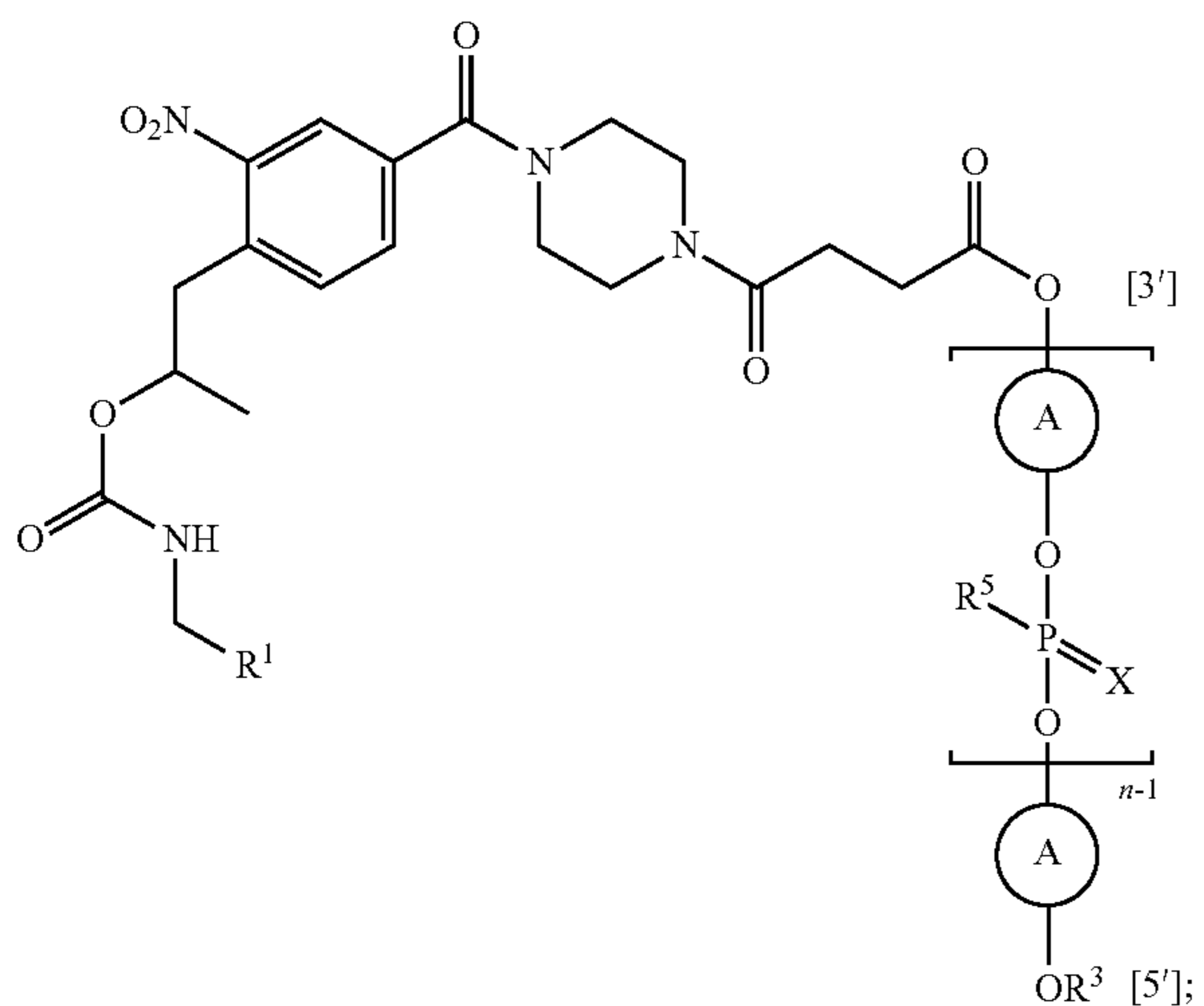
(PG)

(PC)

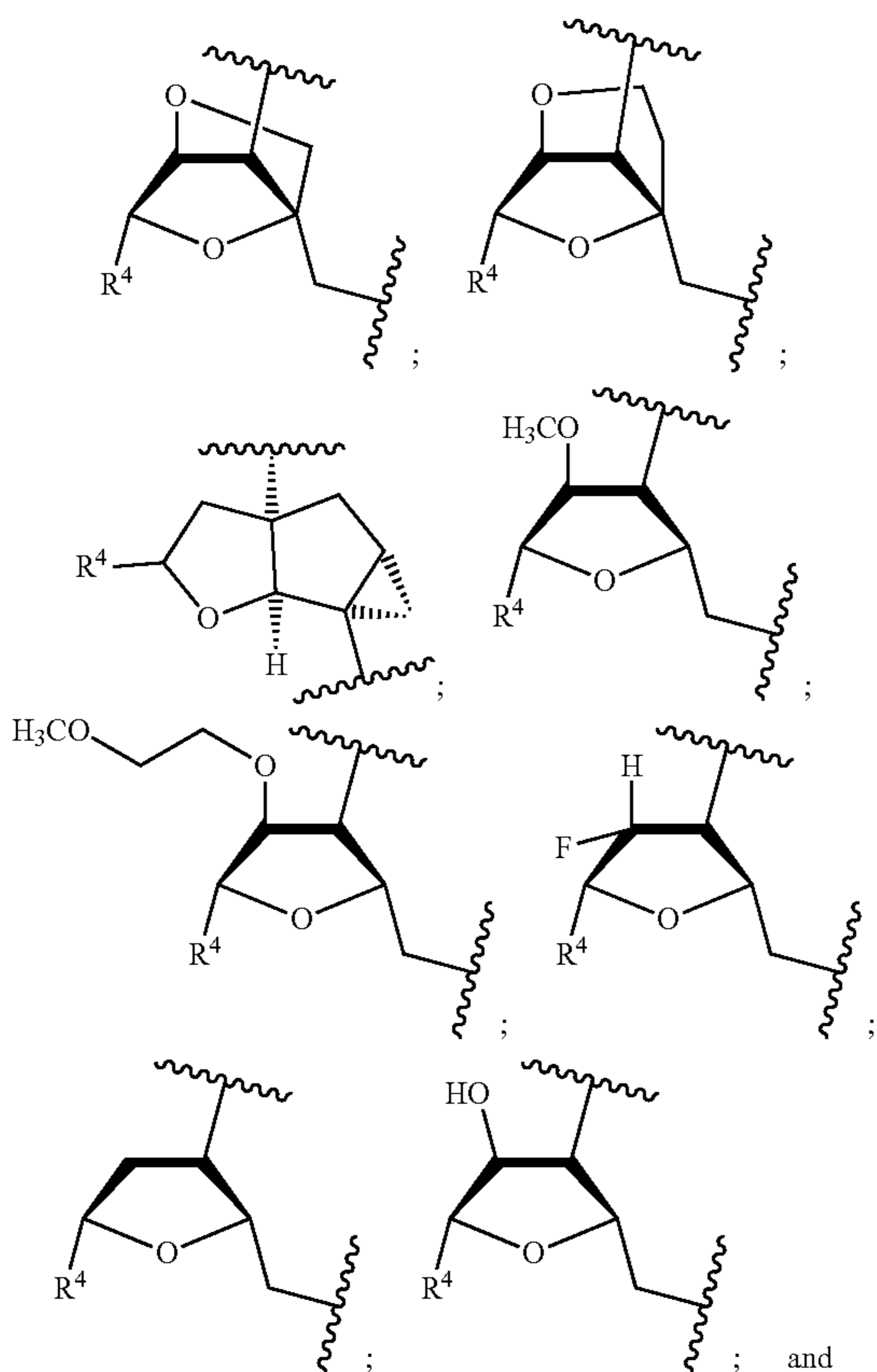
and

209

R³ is, independently for each iteration of (F8), selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and PG is a protecting group,
to form a compound of Formula (F9):

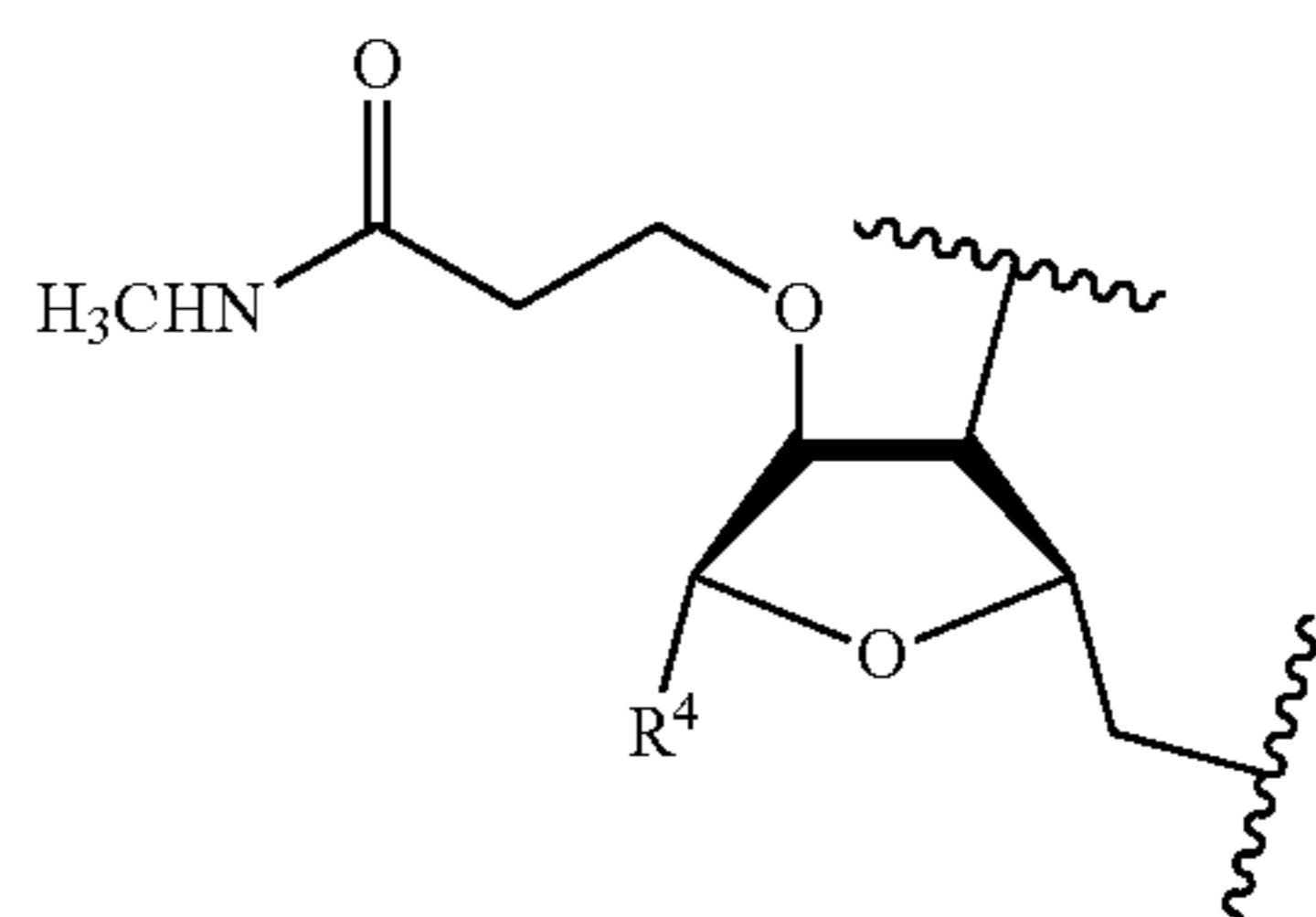


wherein n is an integer from 10 to 40, R¹ is a support-medium, X is O or S, R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN, each A, independently at each occurrence, is selected from the group consisting of:

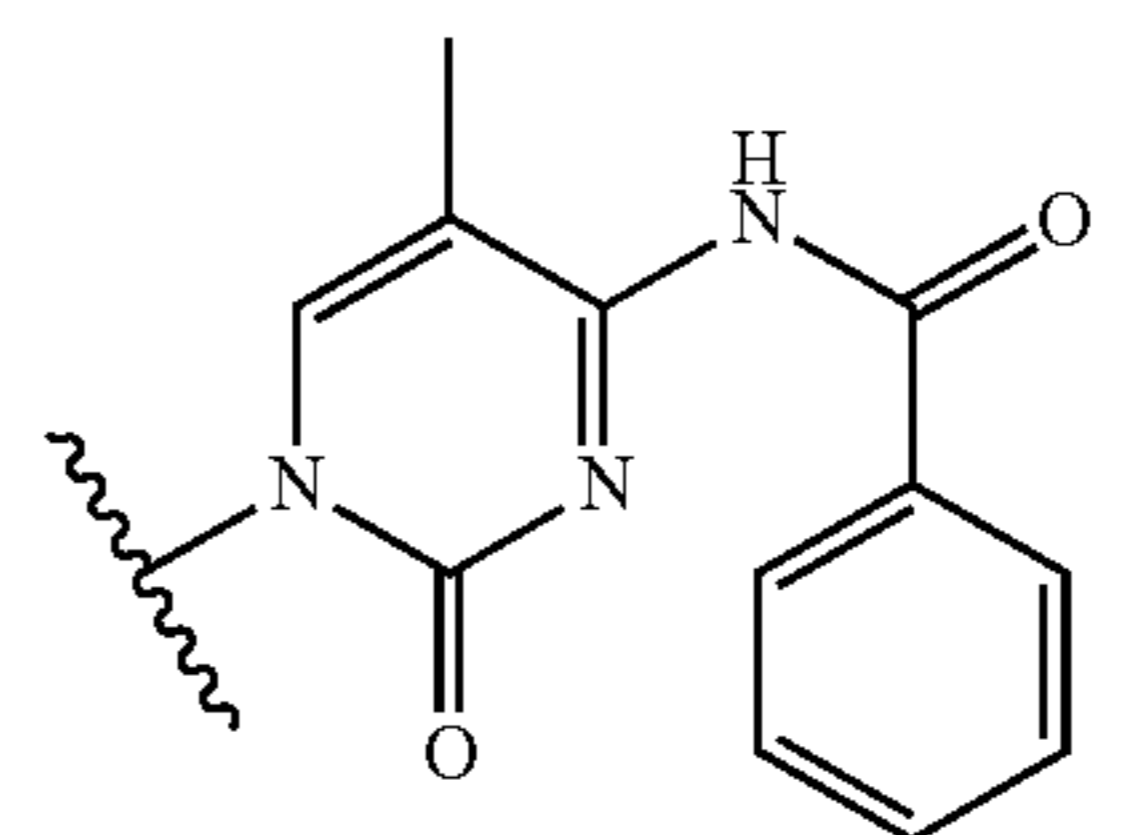
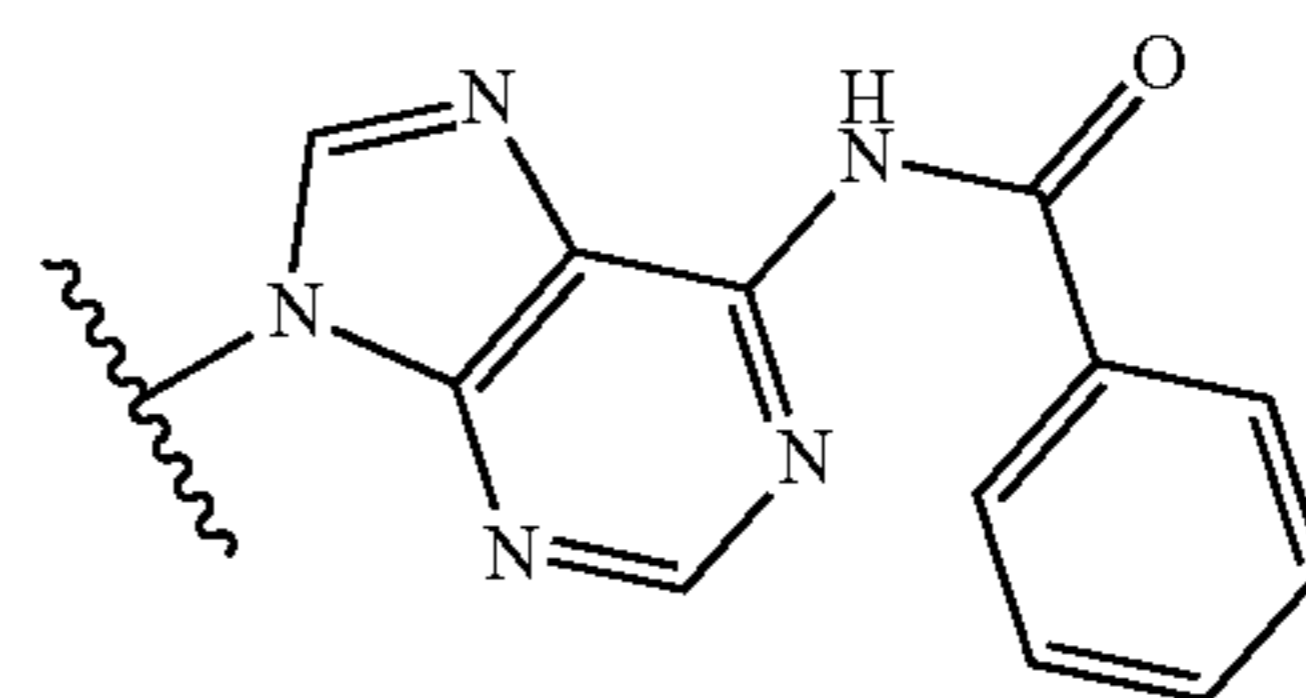
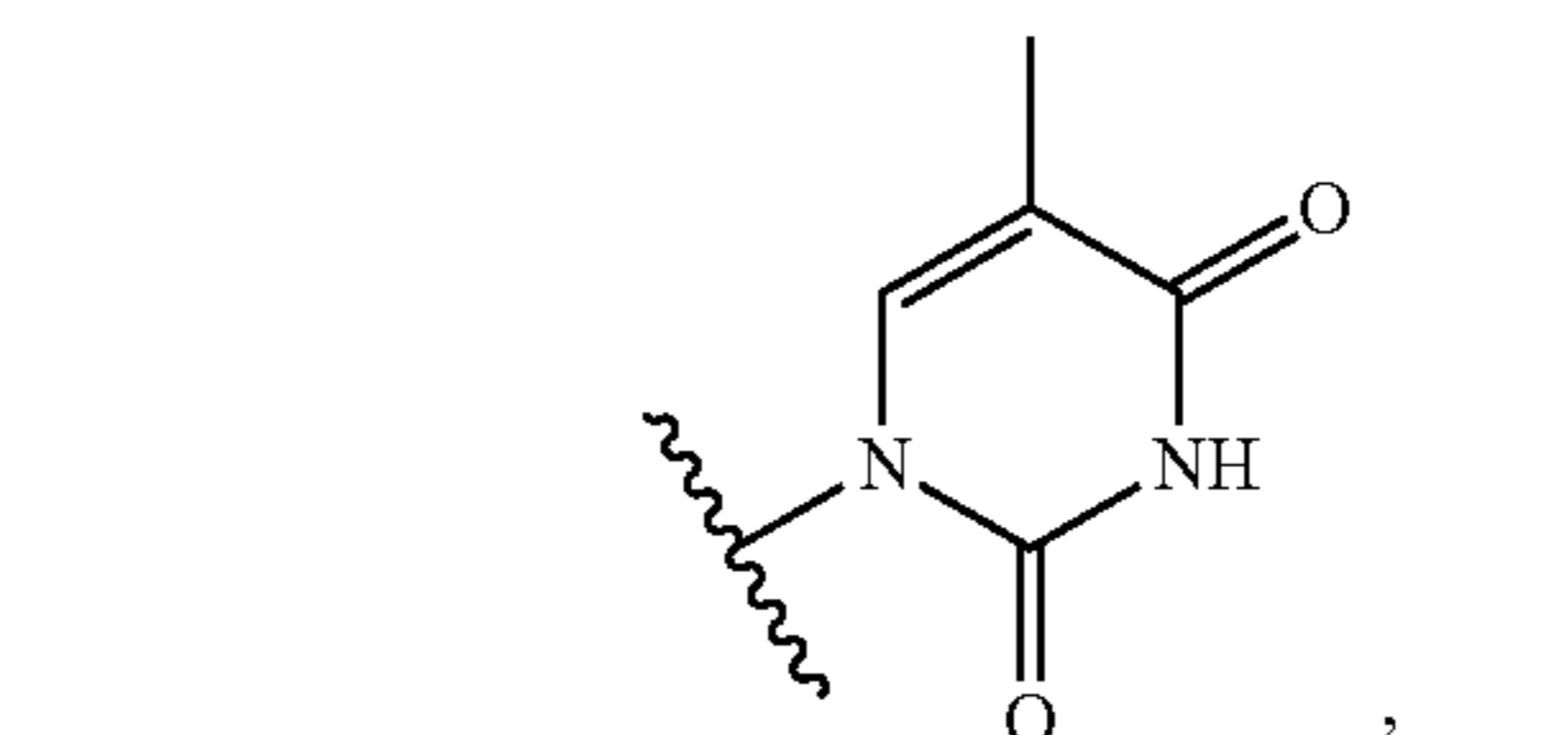
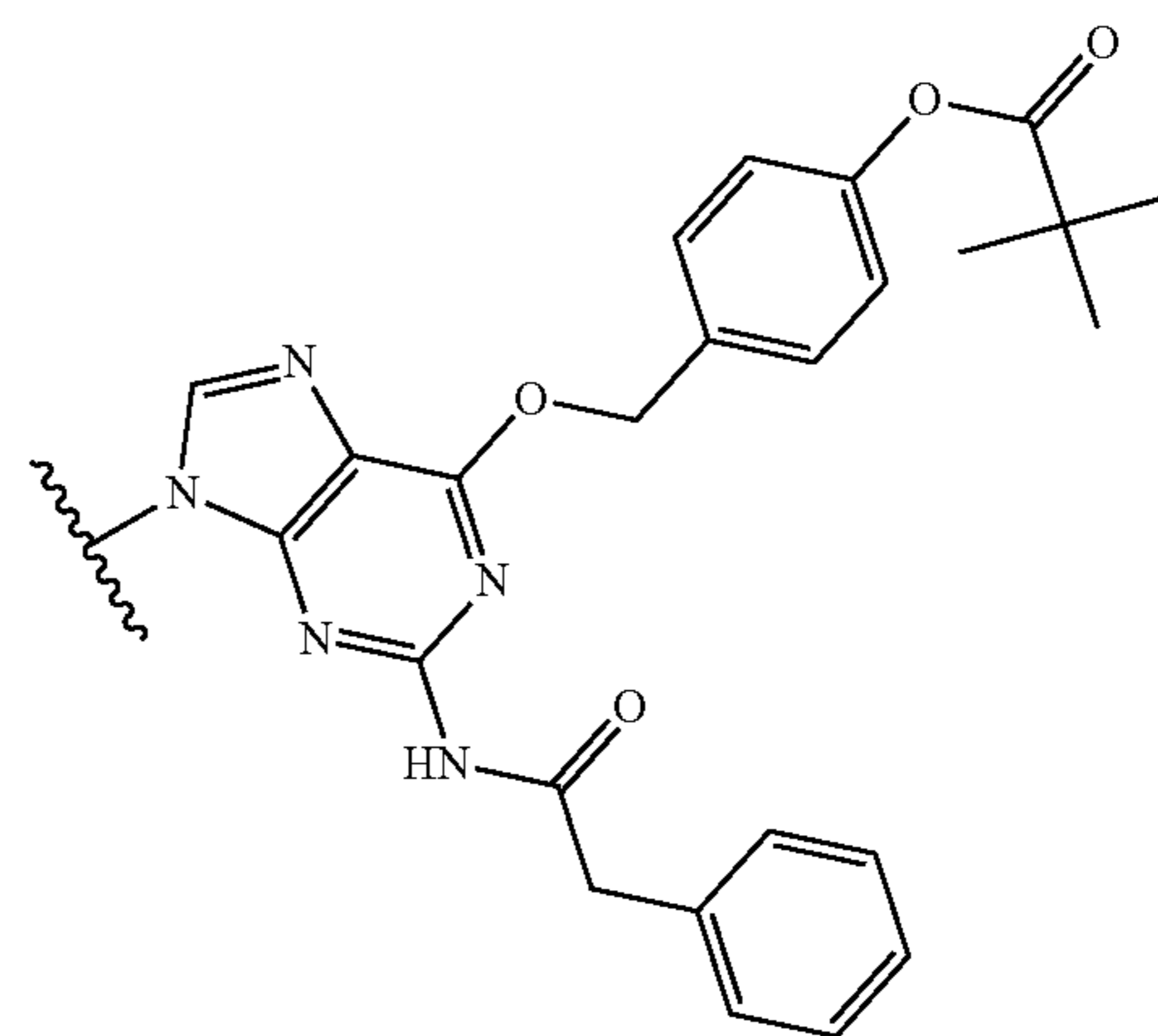
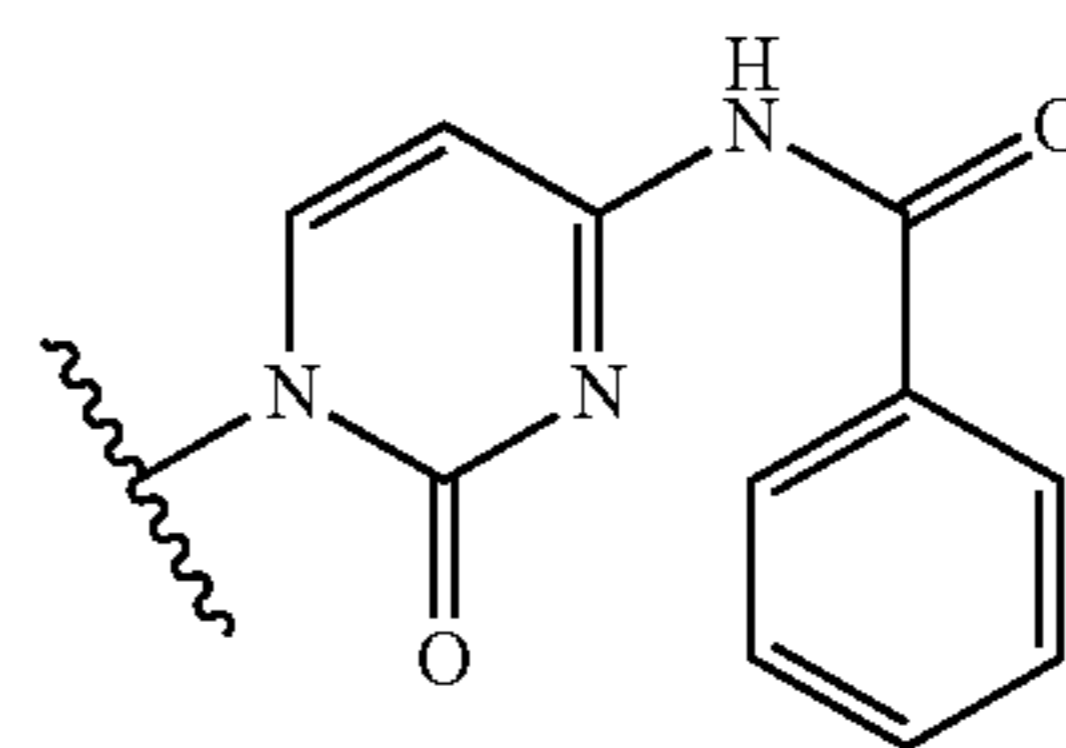


210

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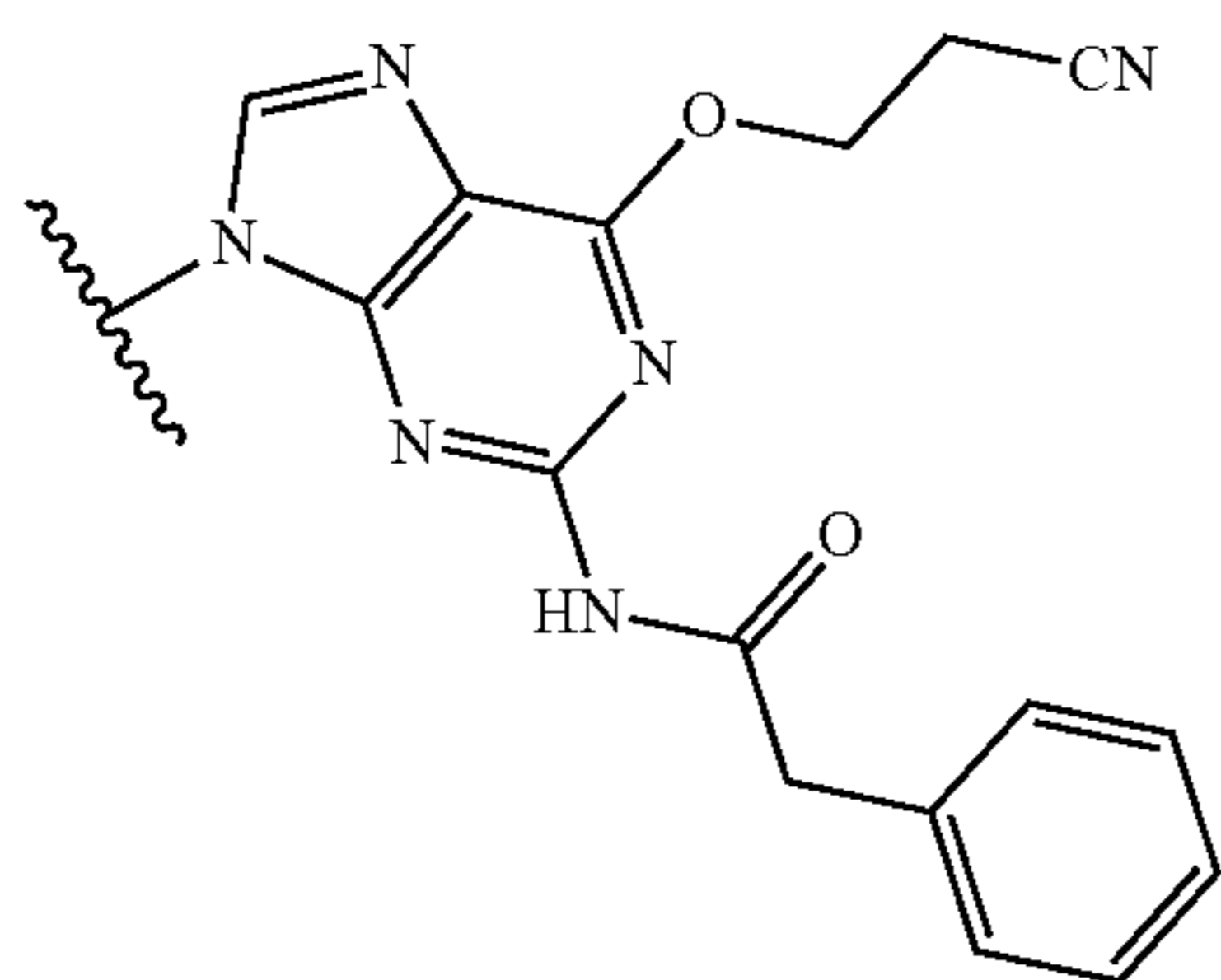
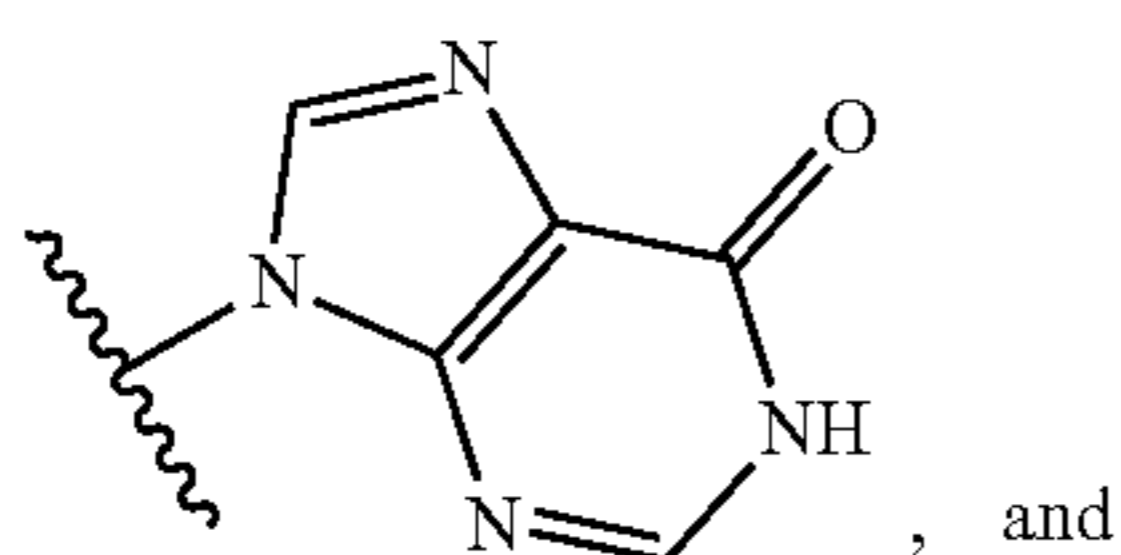
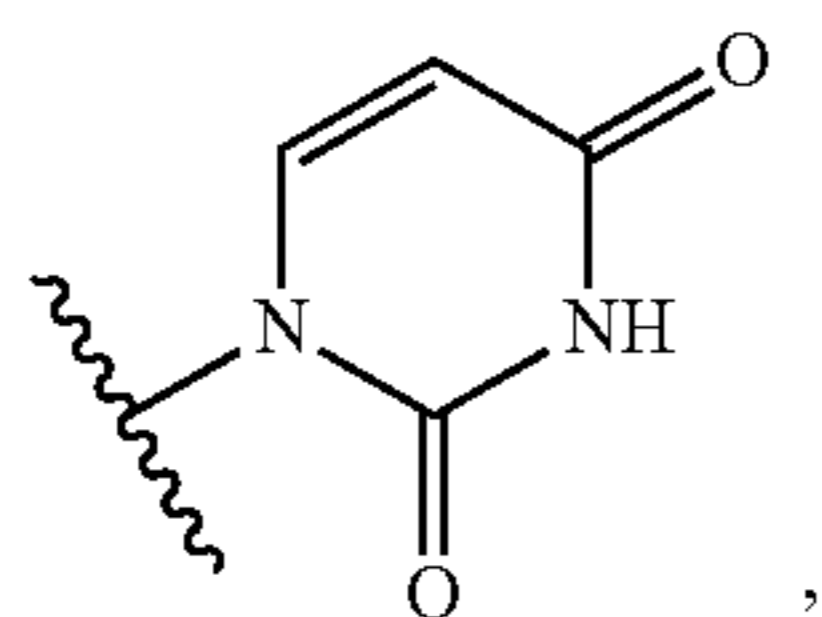


R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each occurrence, selected from the group consisting of:



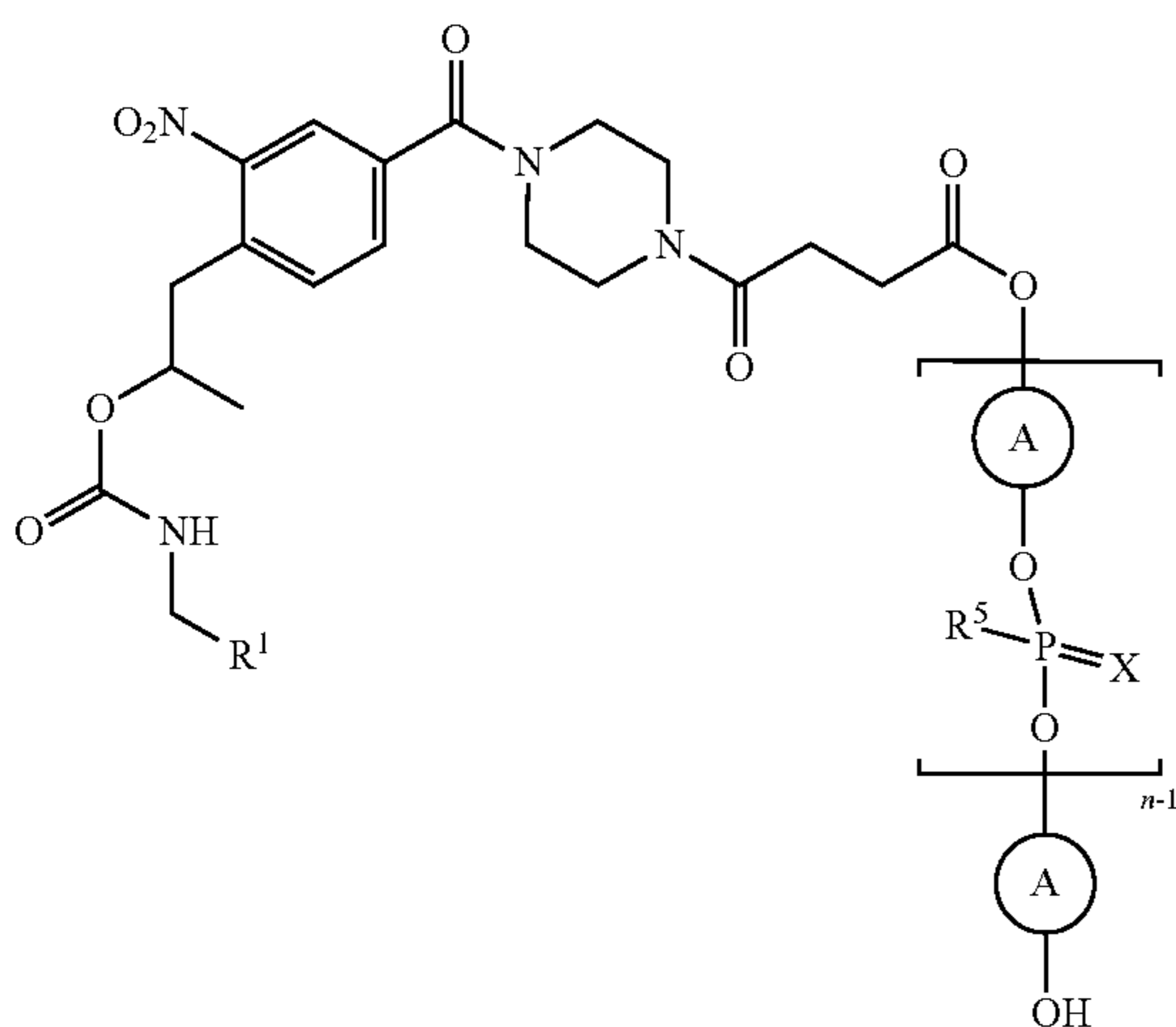
211

-continued



wherein where X is S, then each iteration further comprises a sulfurization step, or where X is O, then each iteration further comprises an oxidation step; and

(f) contacting the compound of Formula (F9) with a deblocking agent to form a compound of Formula (F10):

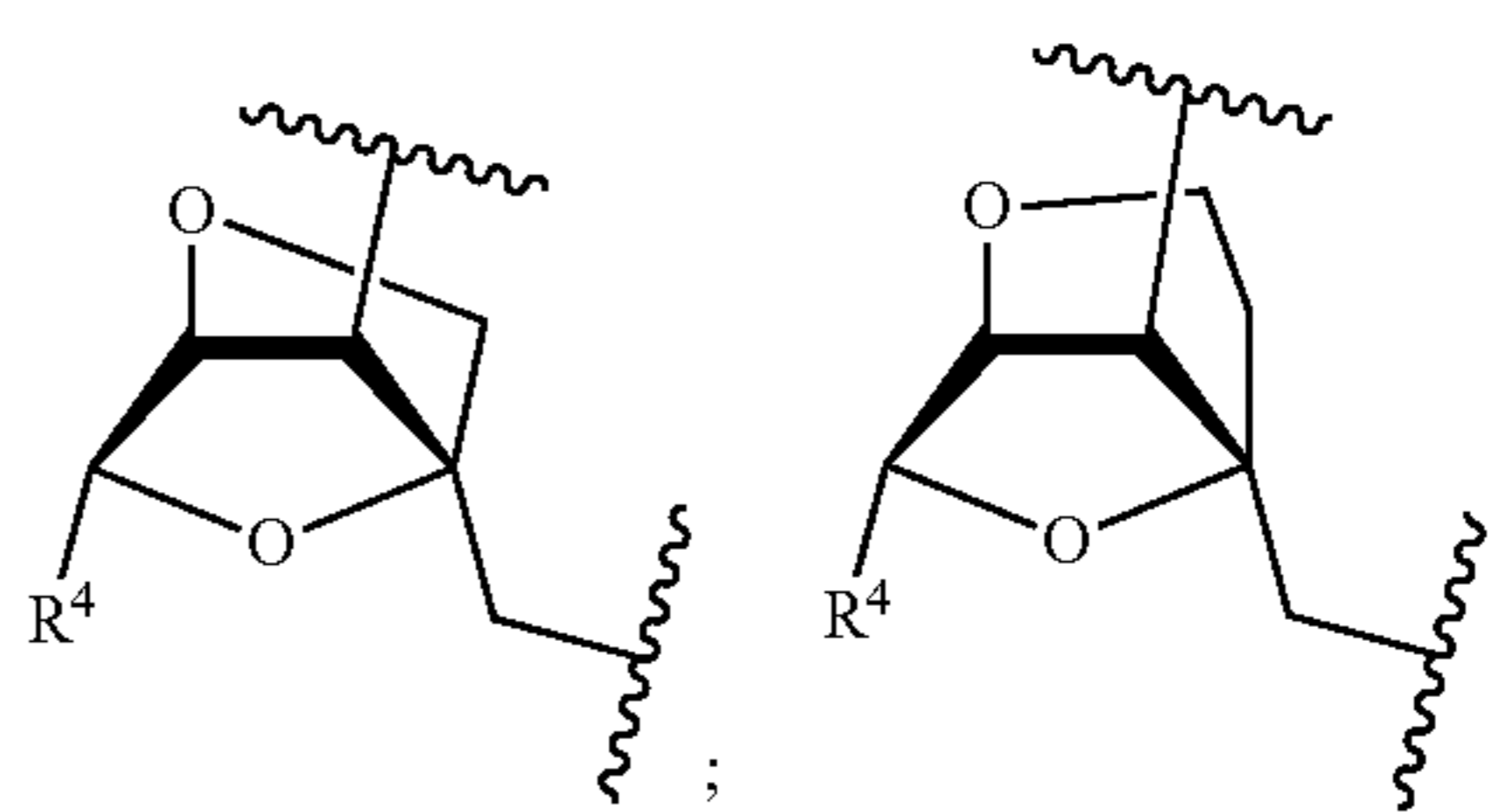


wherein n is an integer from 10 to 40, R¹ is a support-medium, X is O or S, R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN, each A, independently at each occurrence, is selected from the group consisting of:

212

(U)

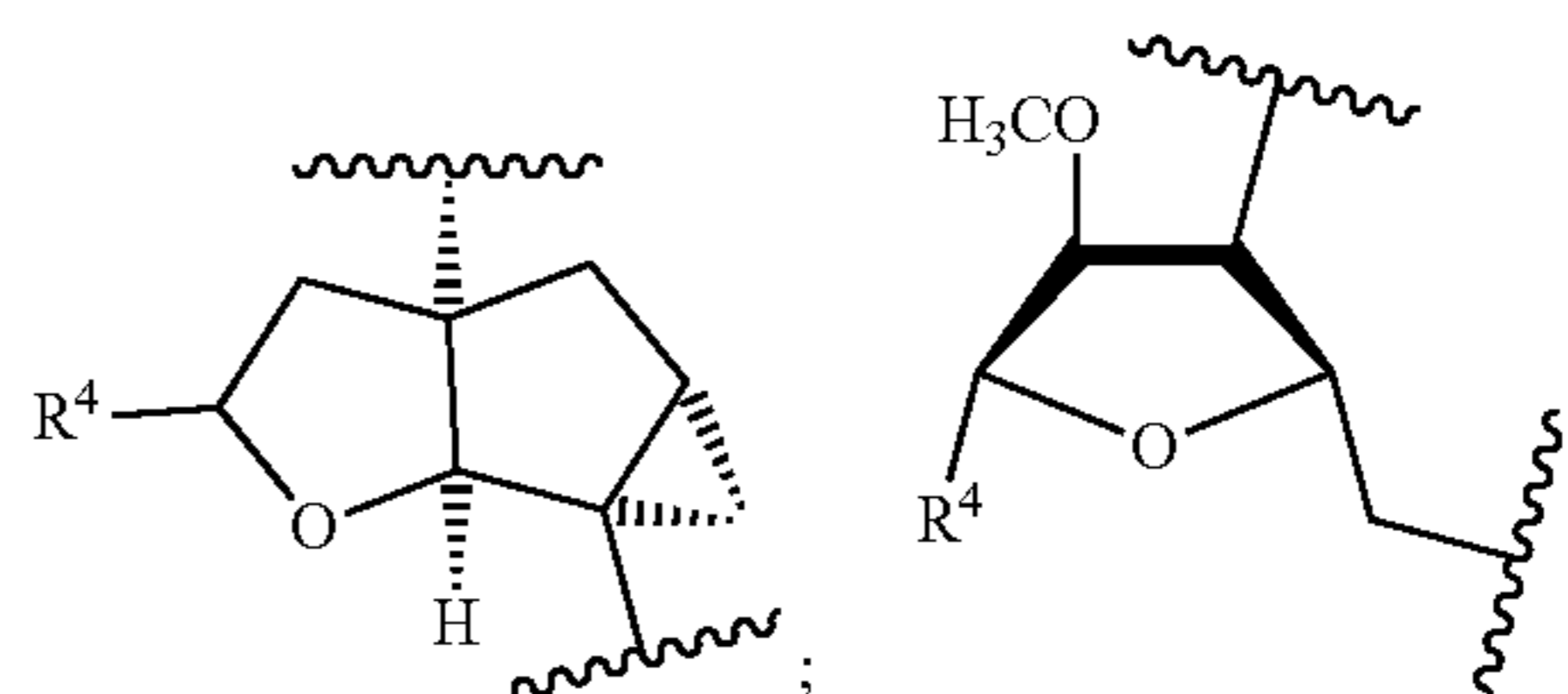
5



10

(I)

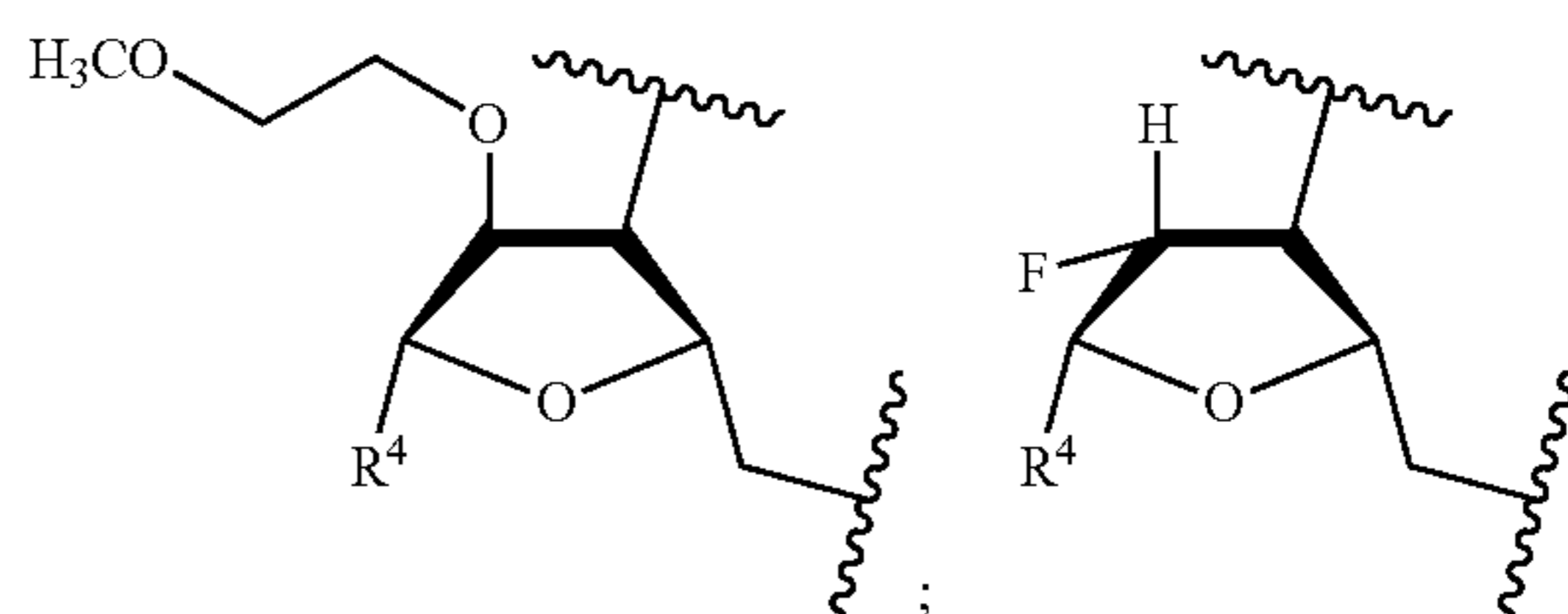
15



20

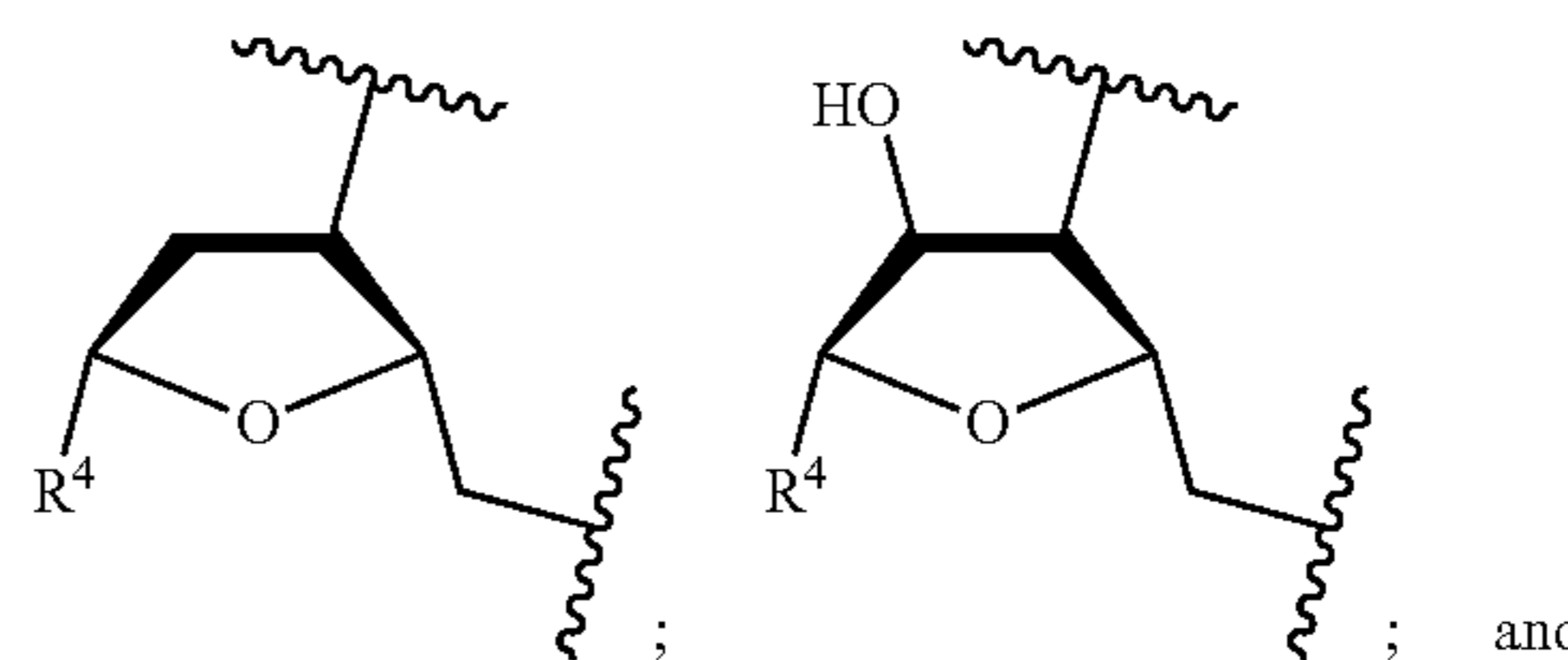
(PG)

25



30

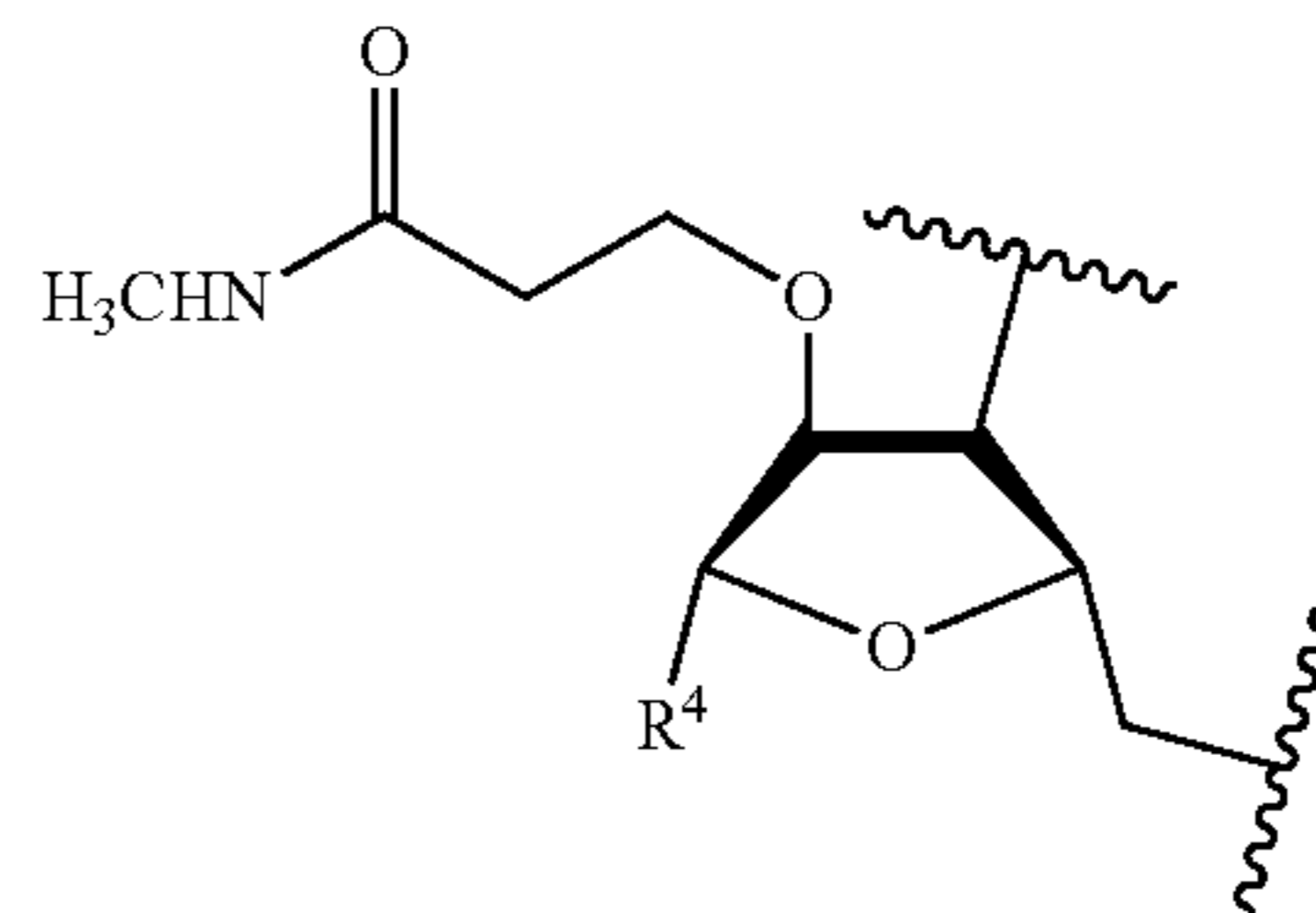
35



40

(F10)

45

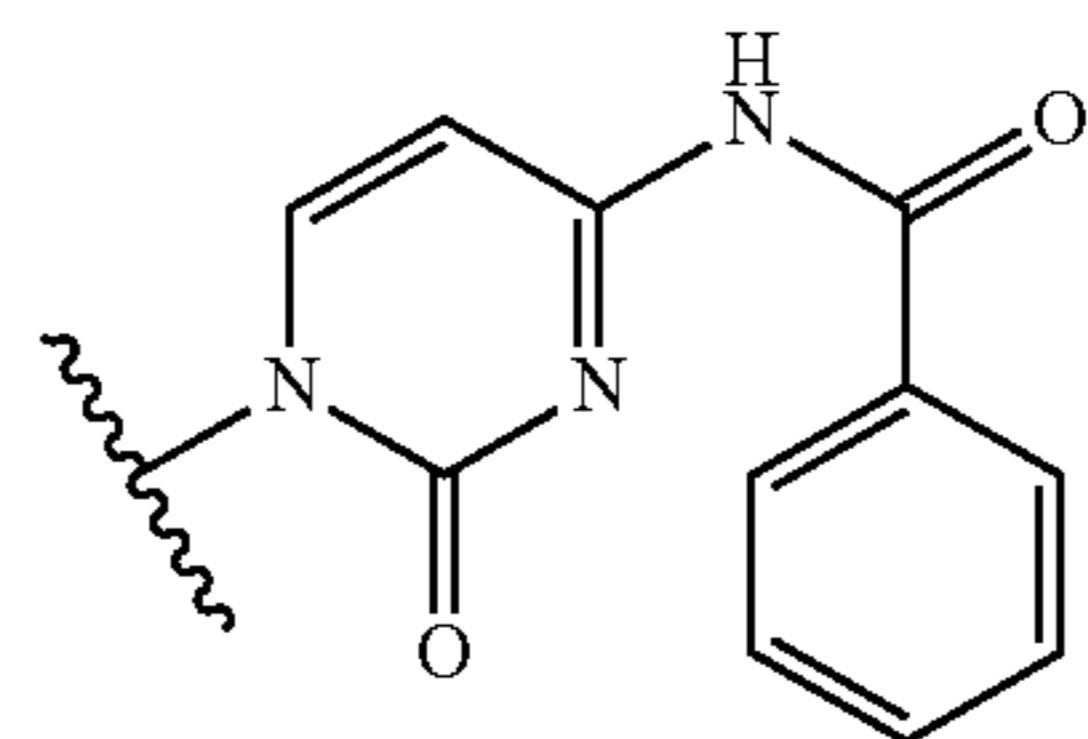


50

55

and R⁴ is, independently for each occurrence, selected from the group consisting of:

60

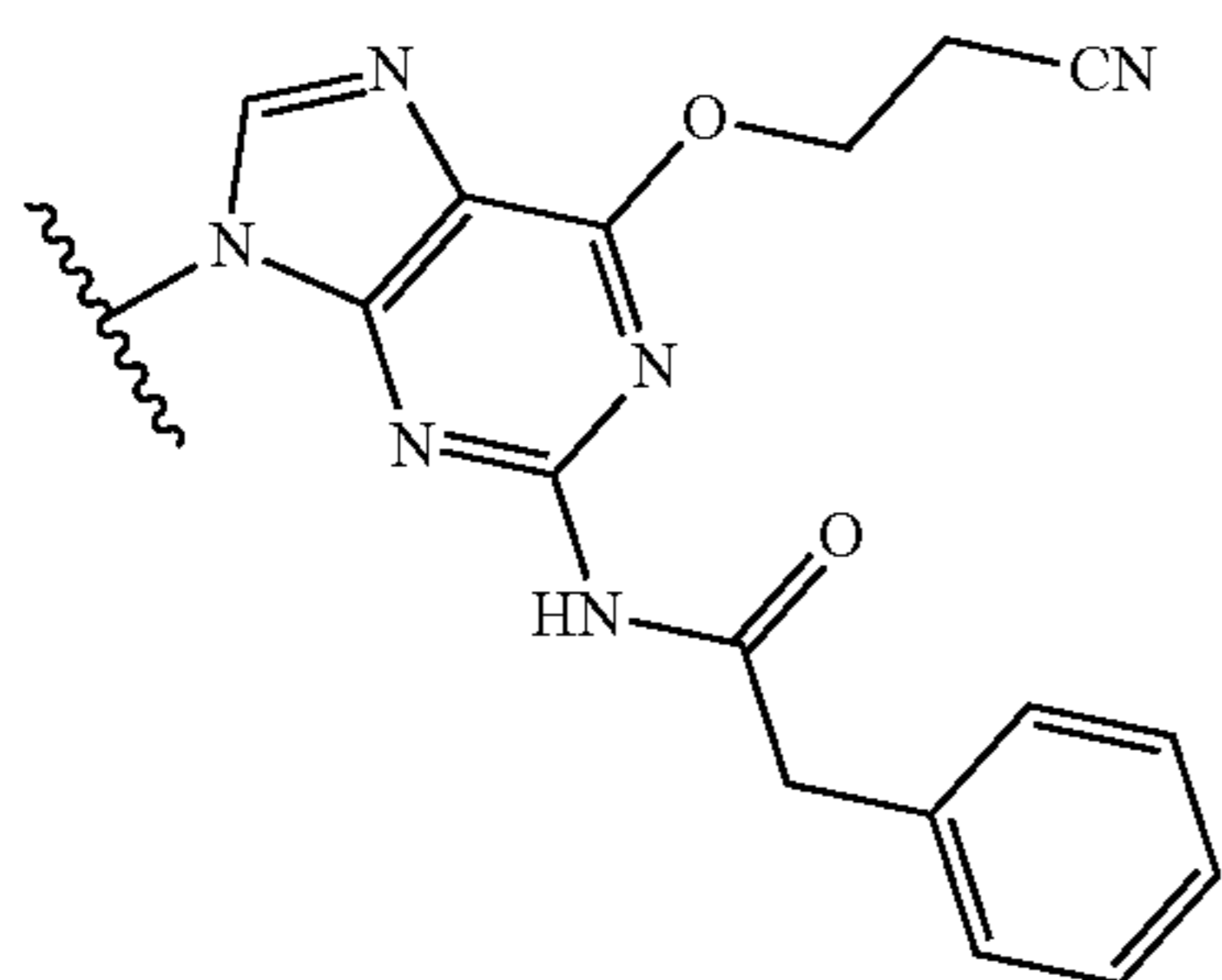
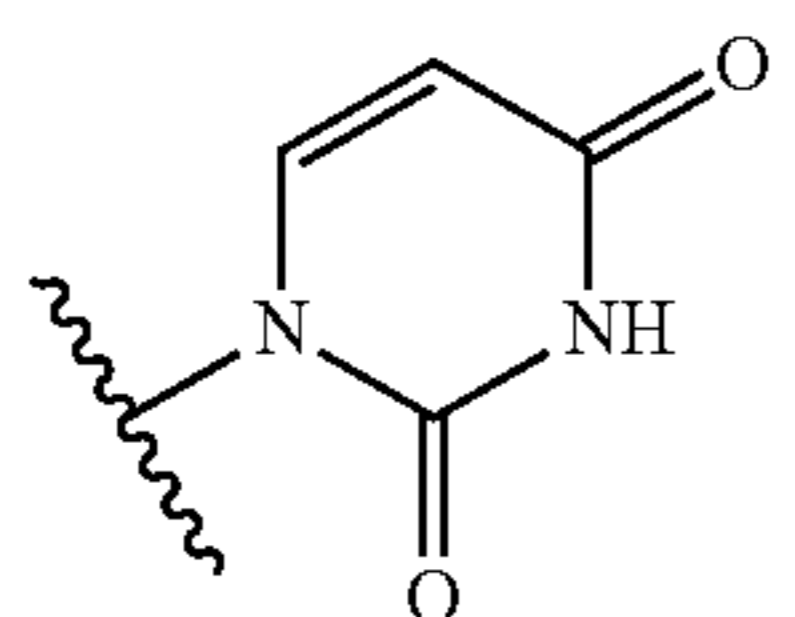
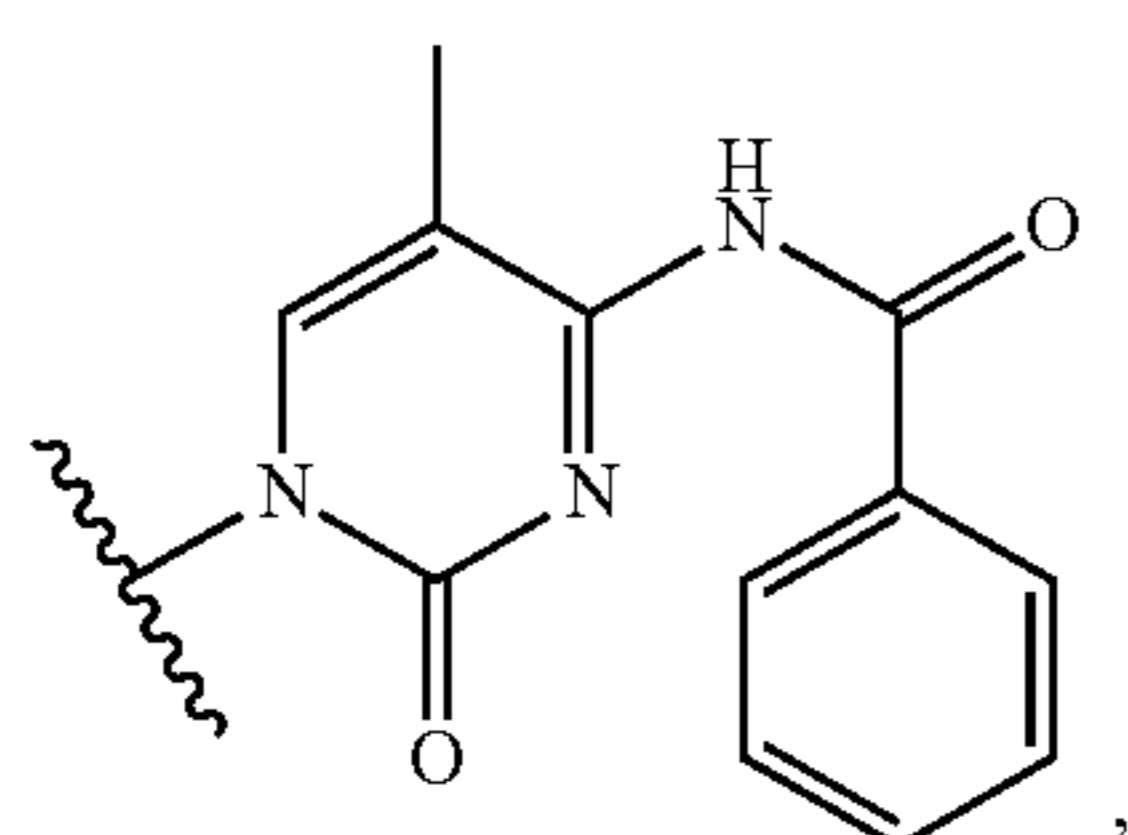
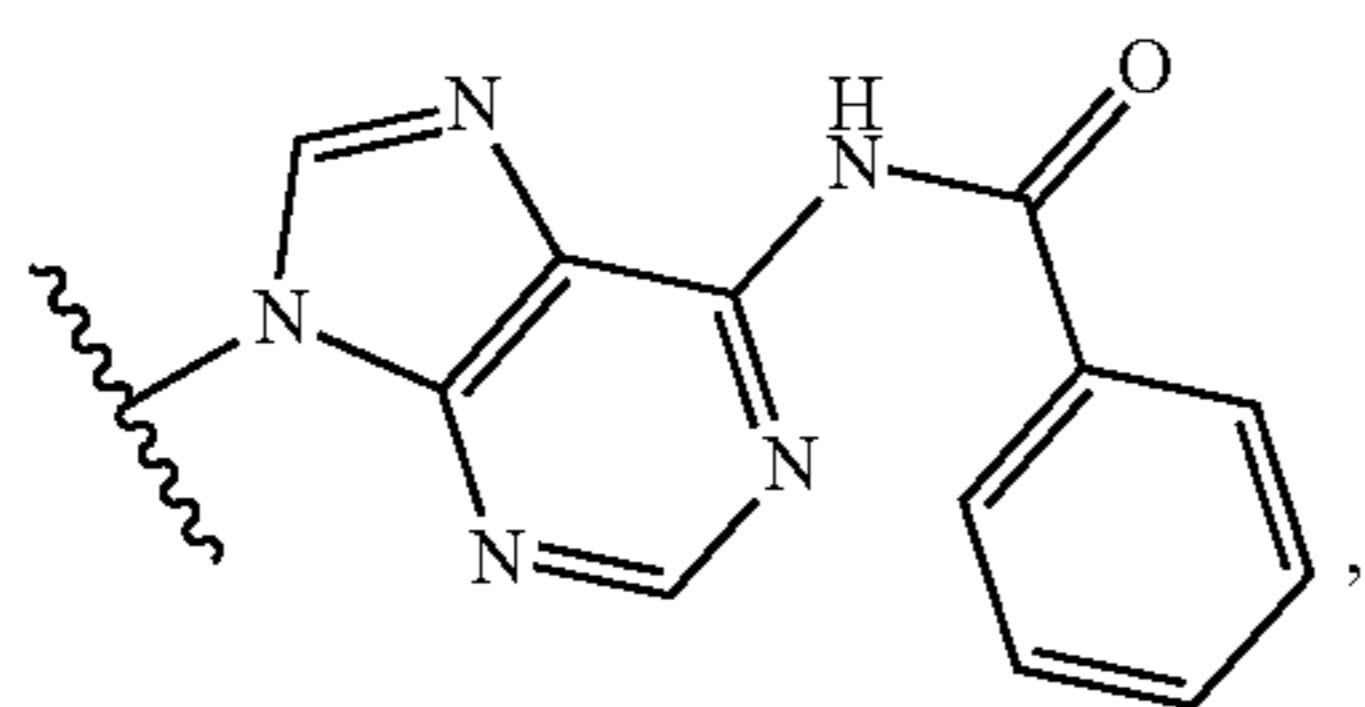
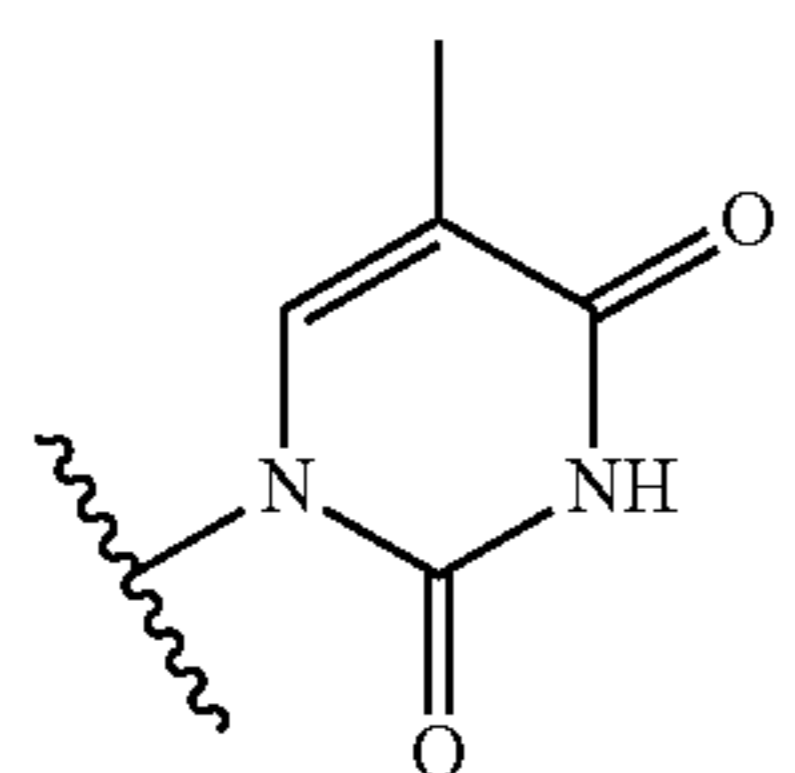
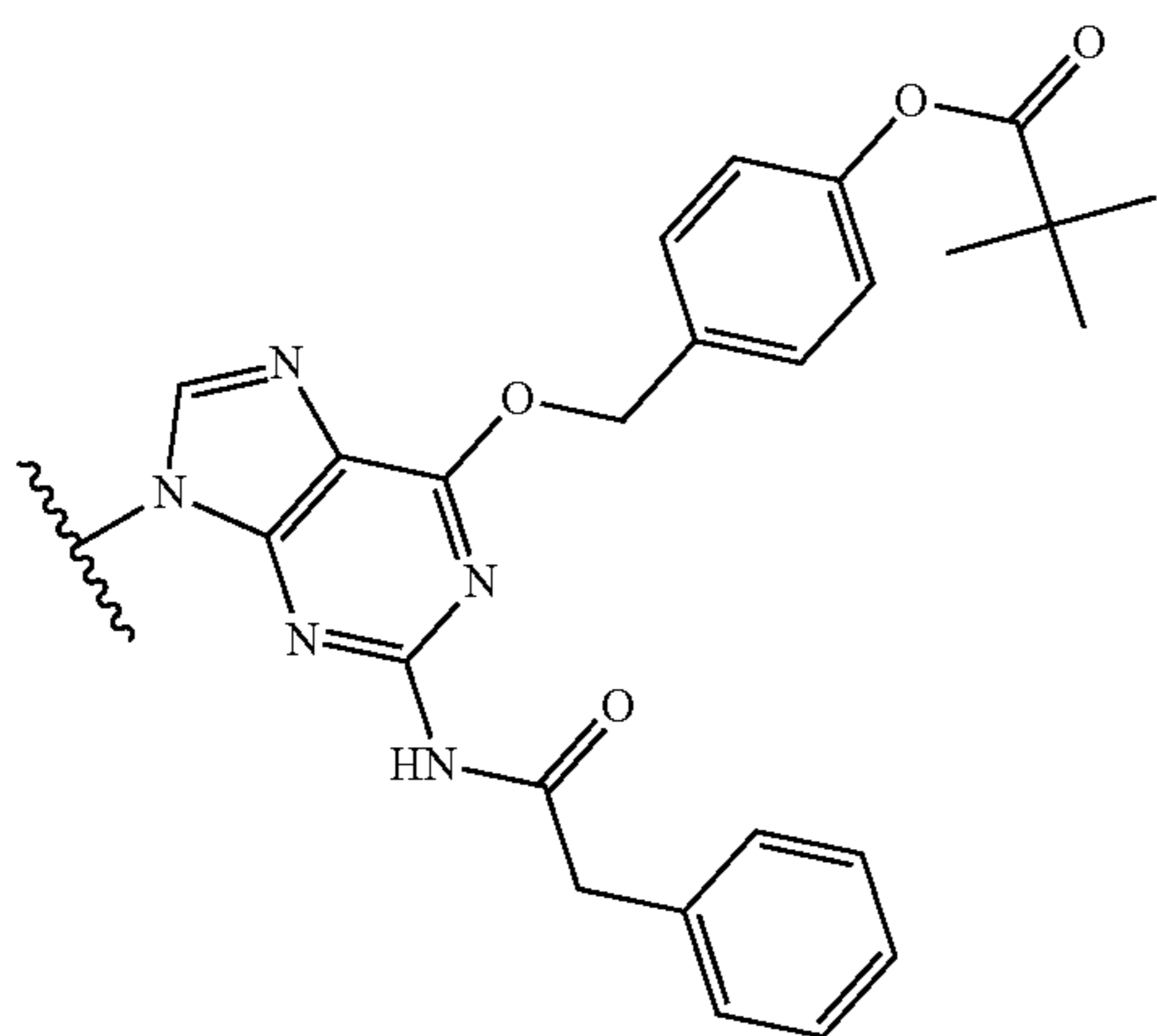


(PC)

65

213

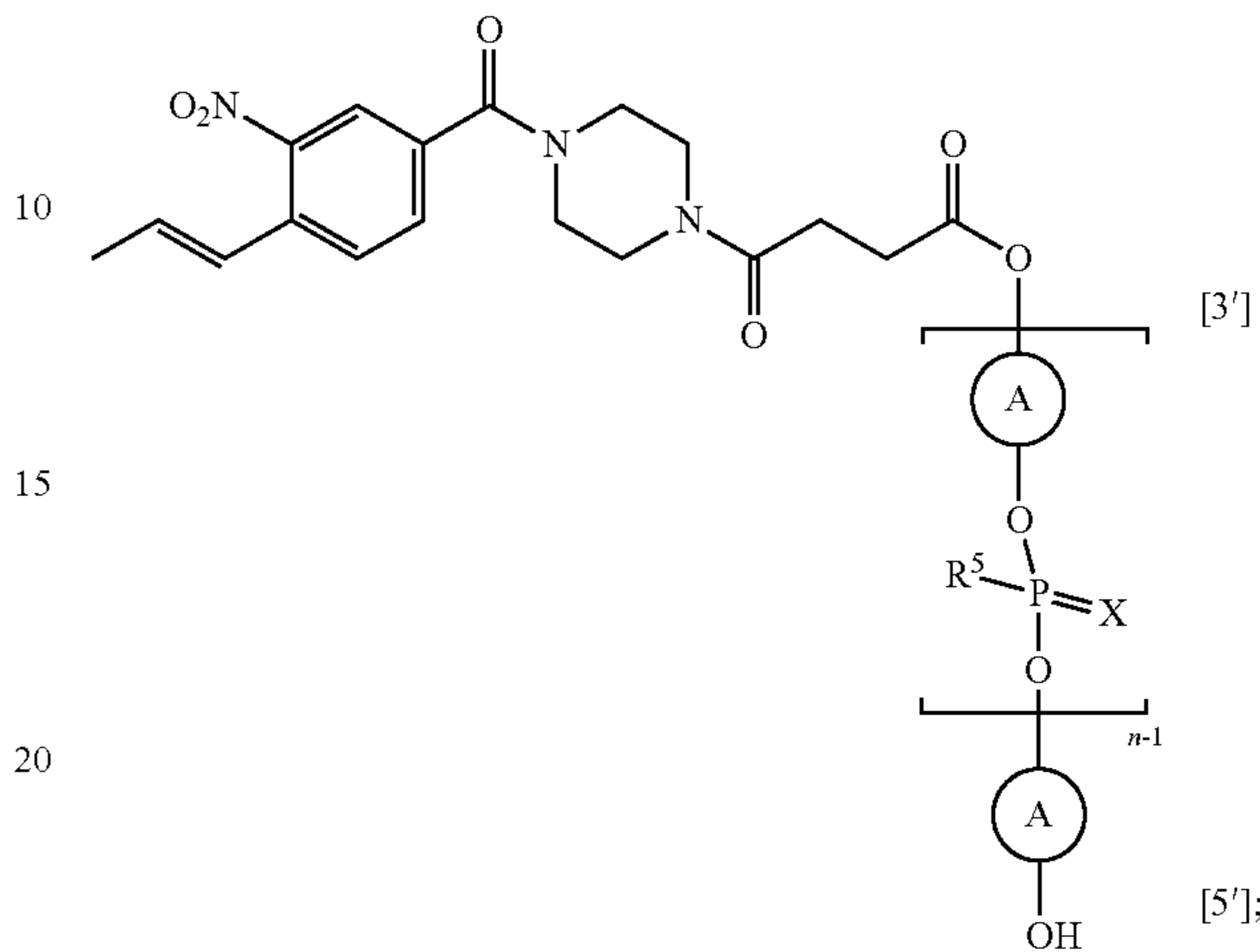
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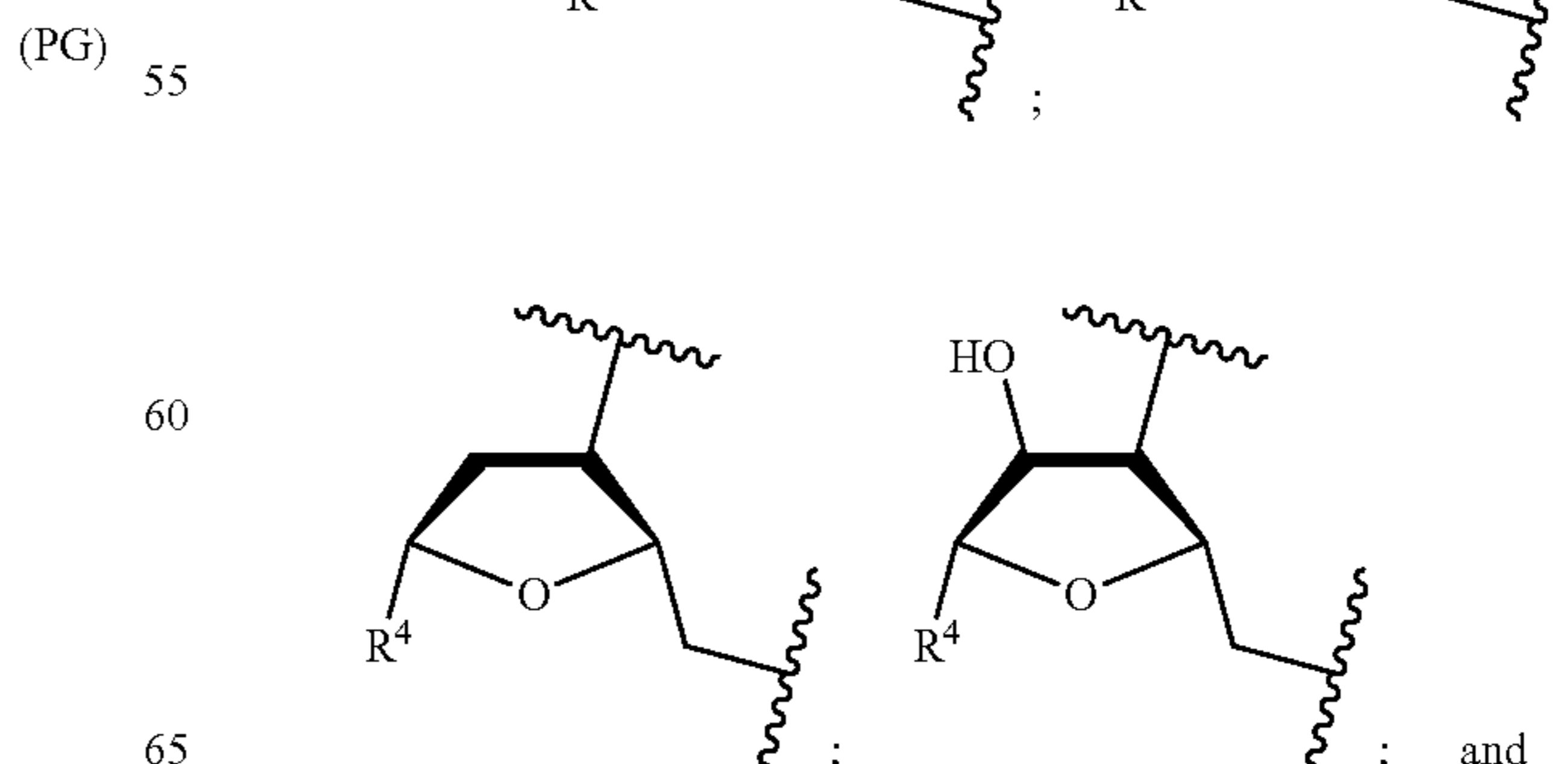
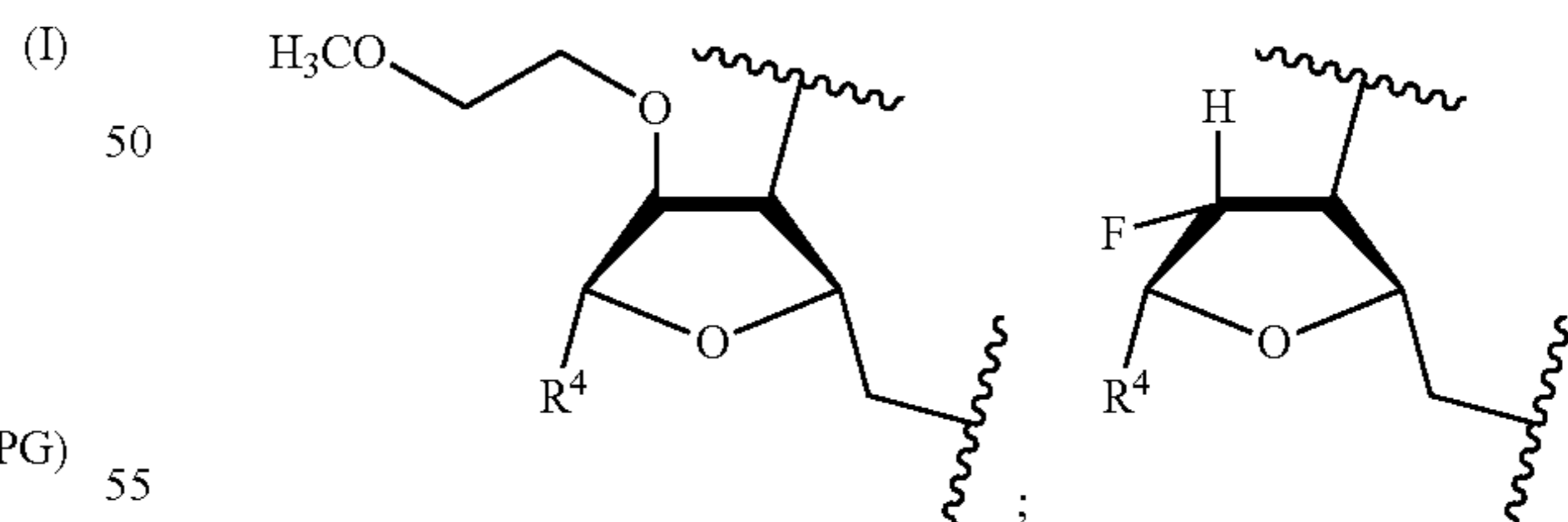
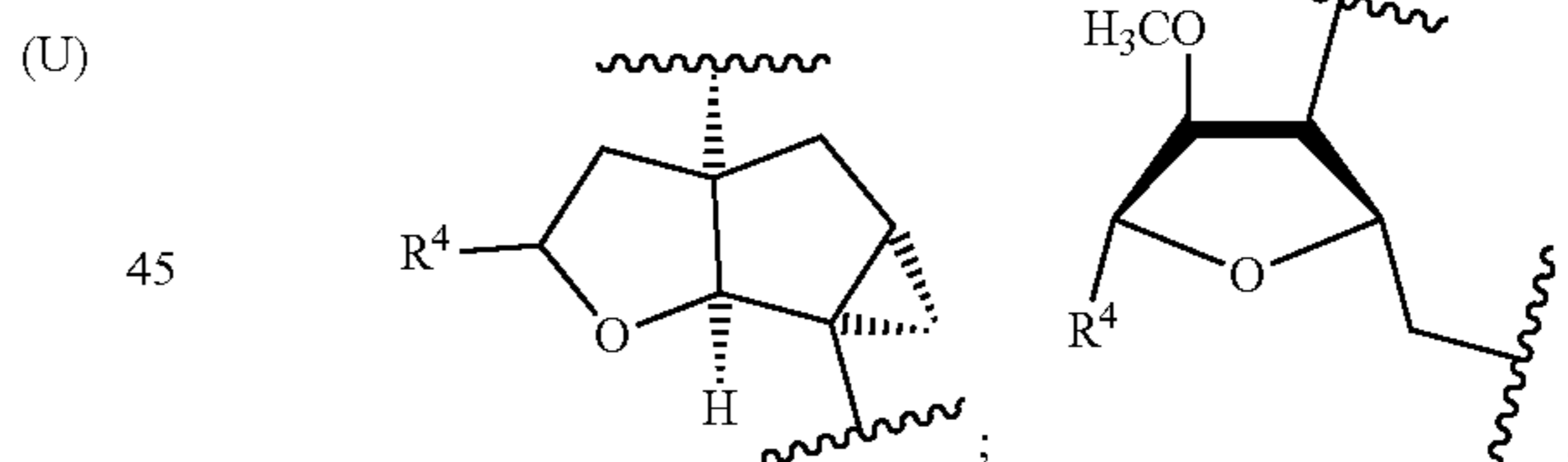
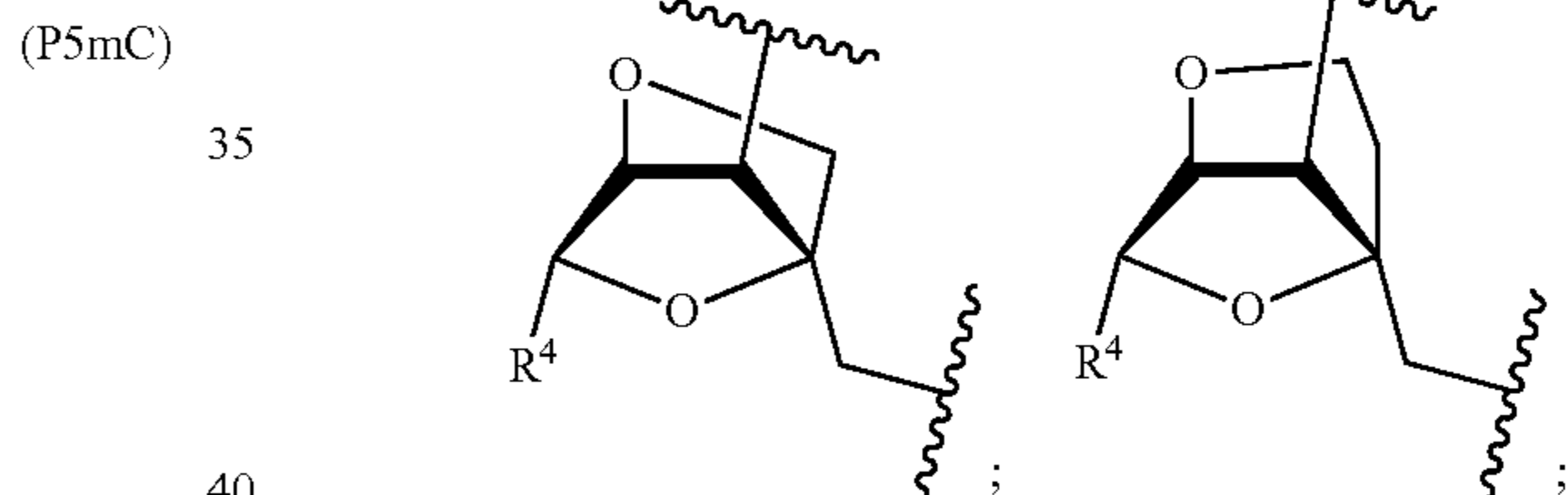
(g) contacting the compound of Formula (F10) with a cleaving agent to form a compound of Formula (F11):

5 (F11)



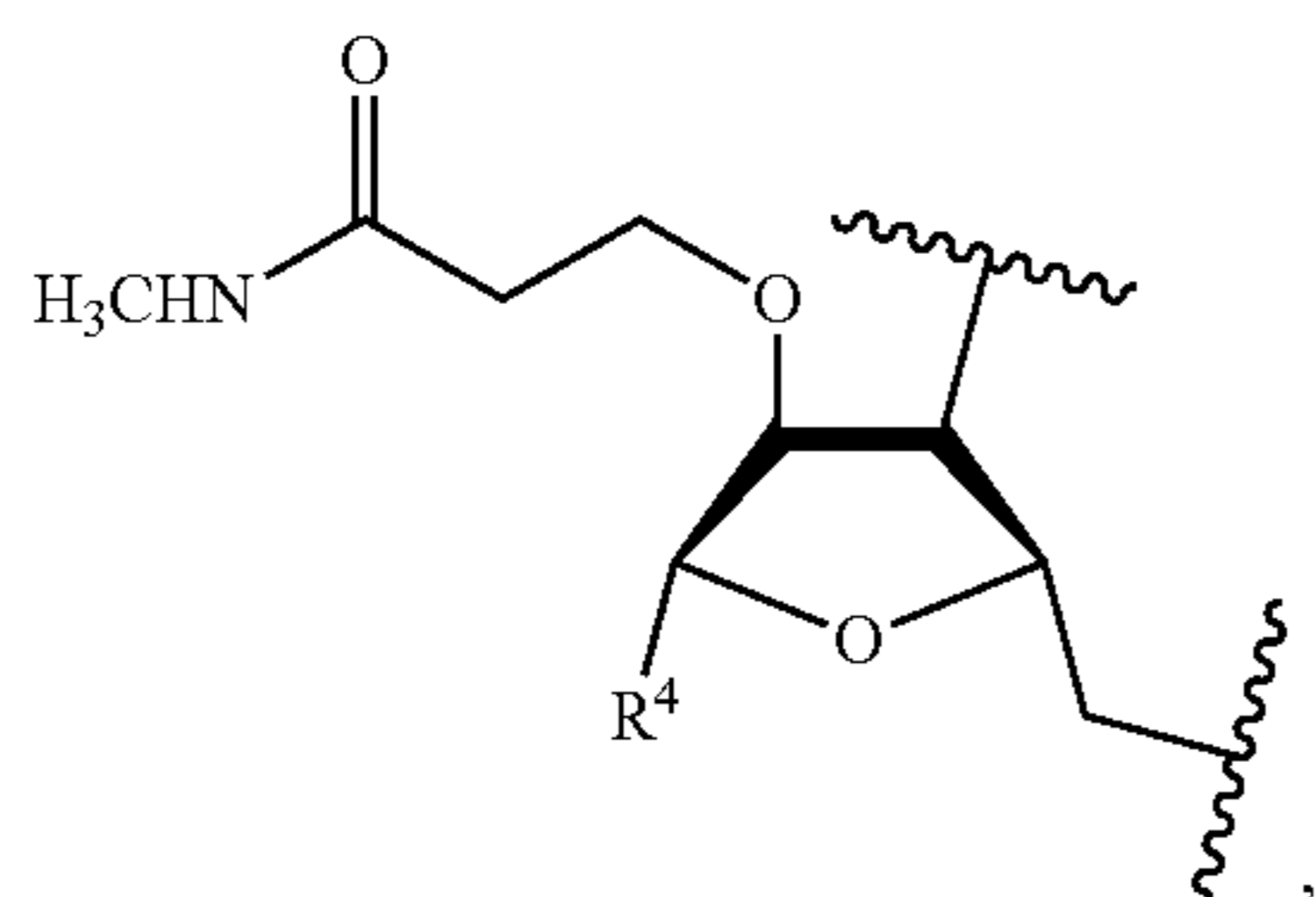
25 wherein n is an integer from 10 to 40,

X is O or S, R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN, each A, independently at each occurrence, is selected from the group consisting of:

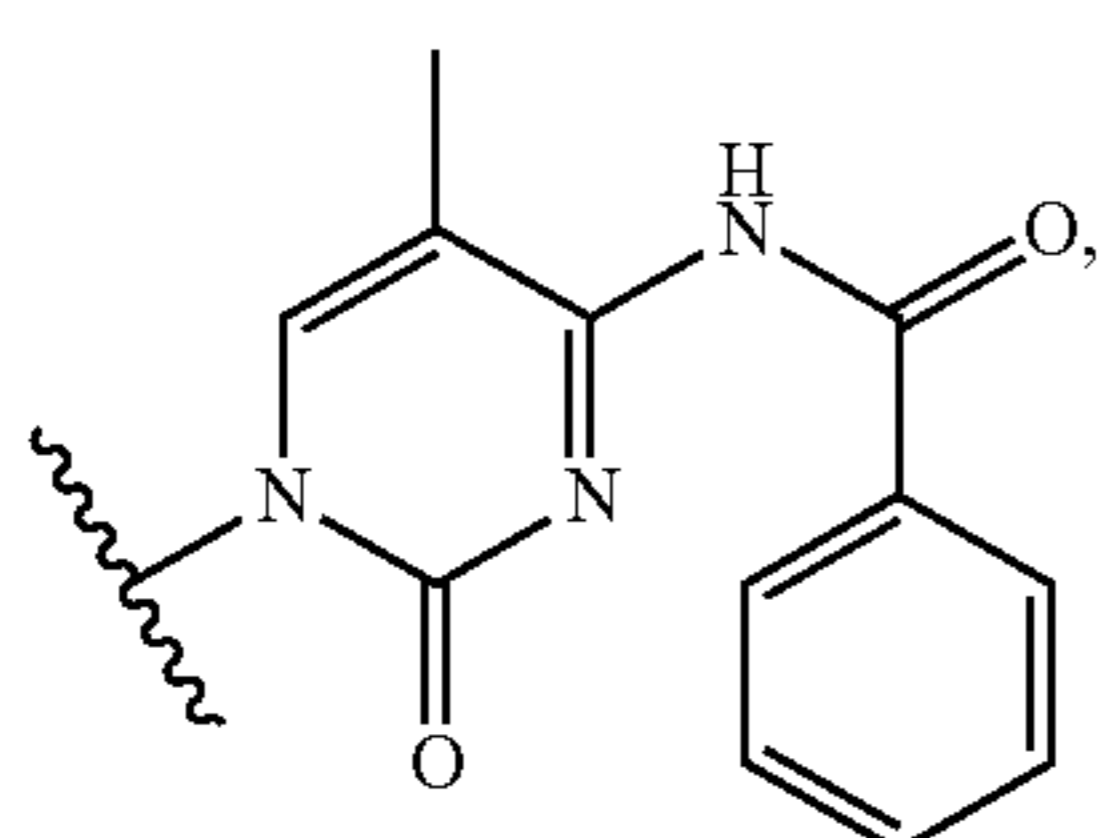
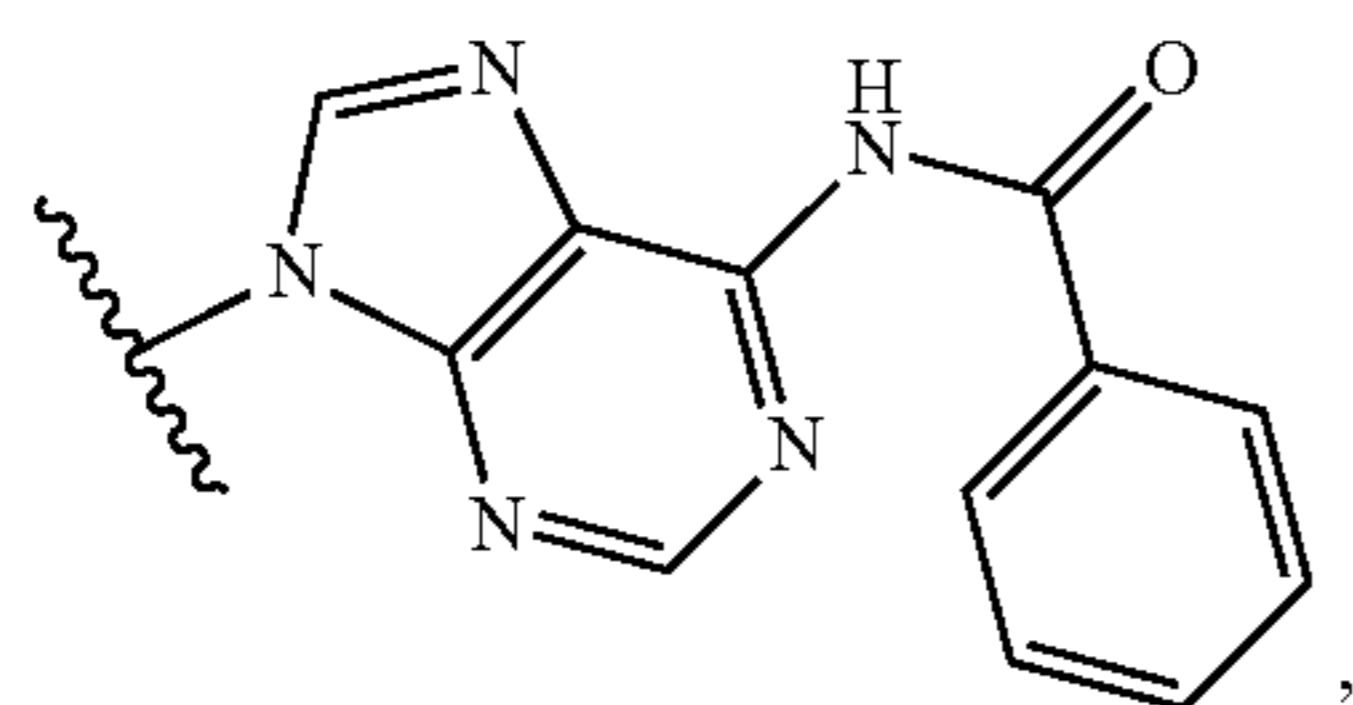
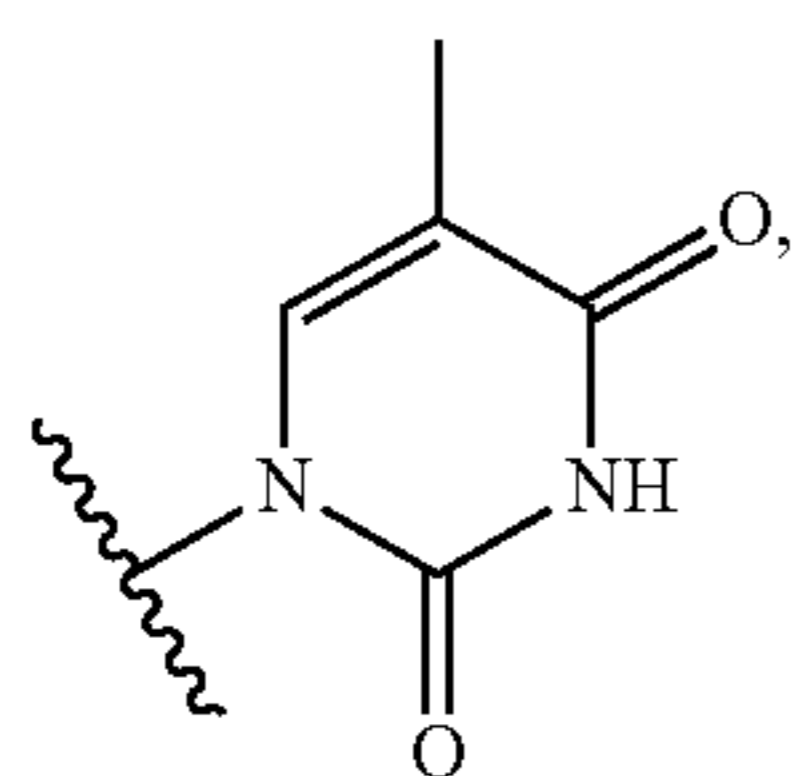
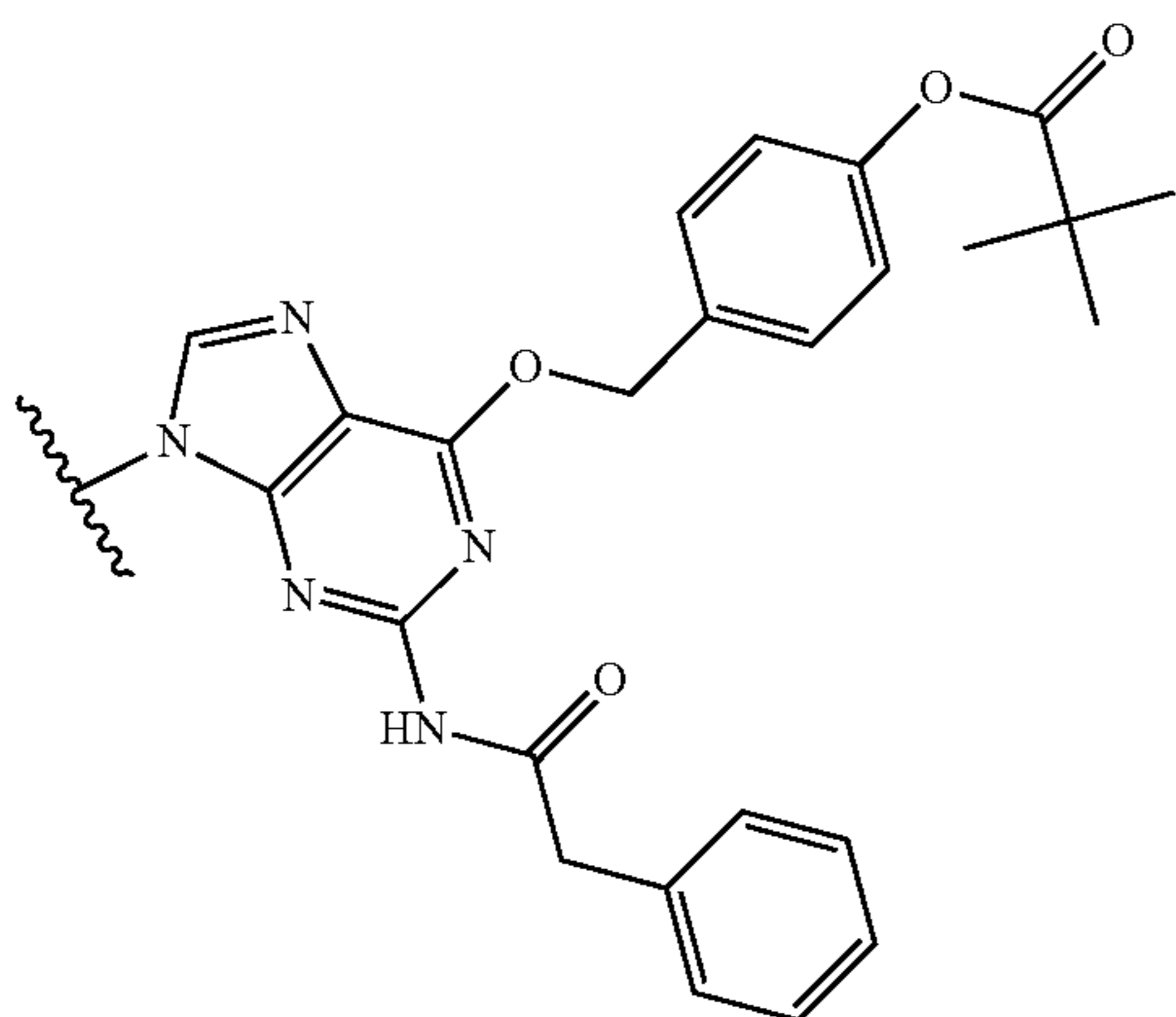
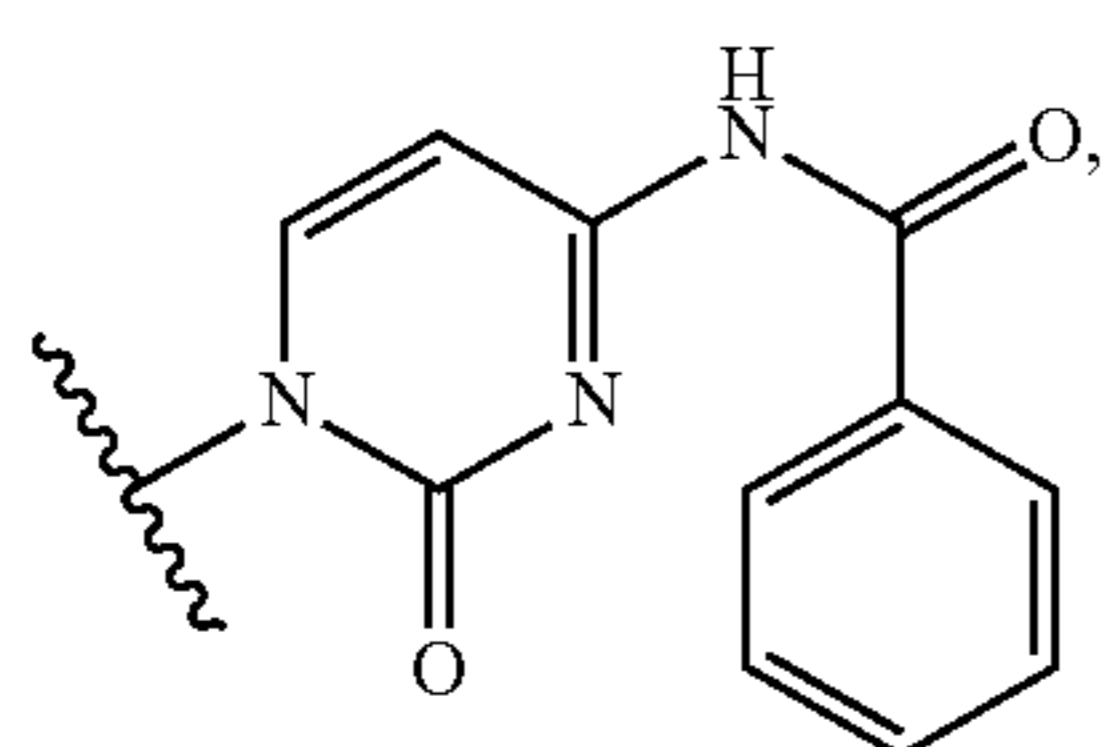


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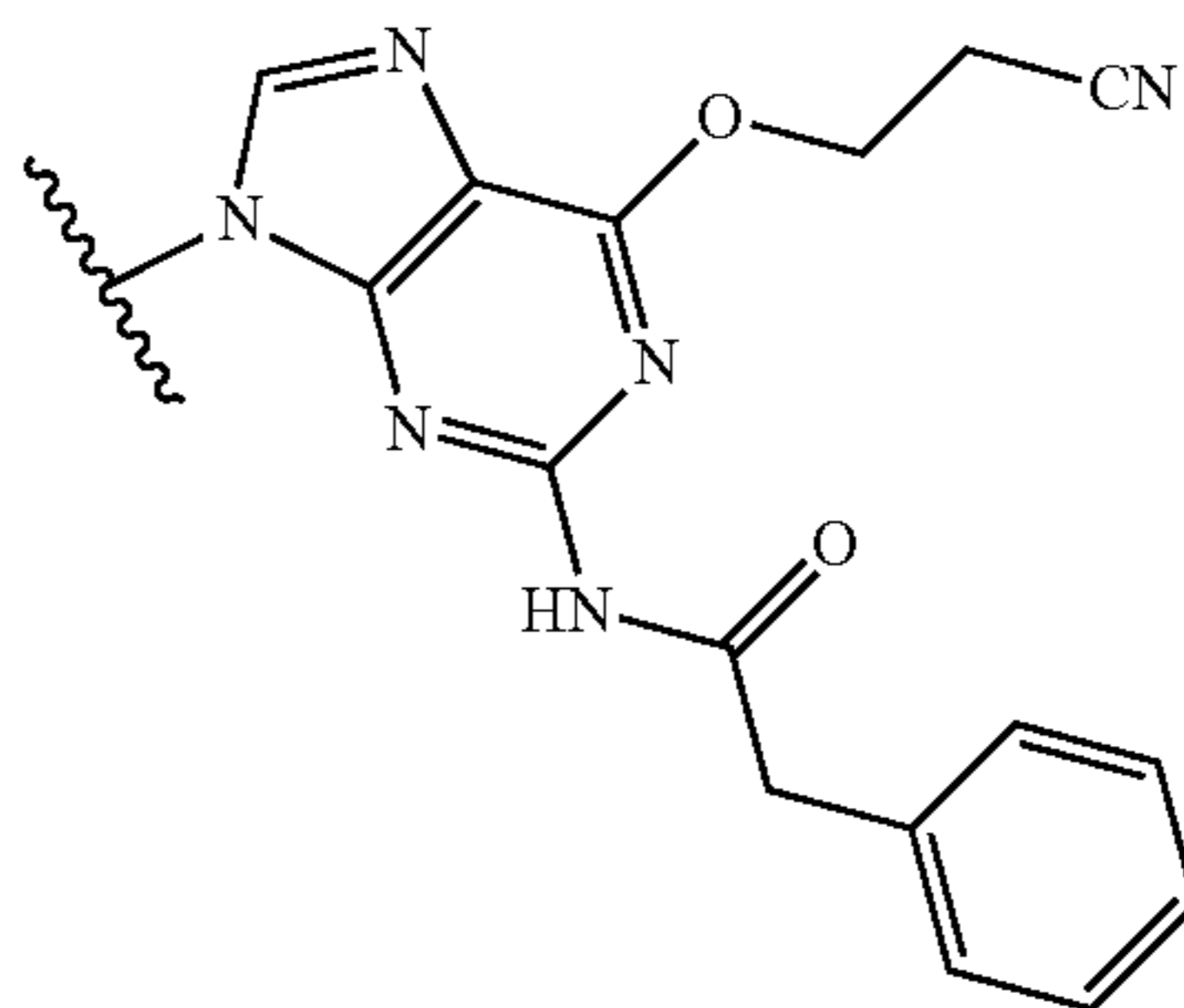
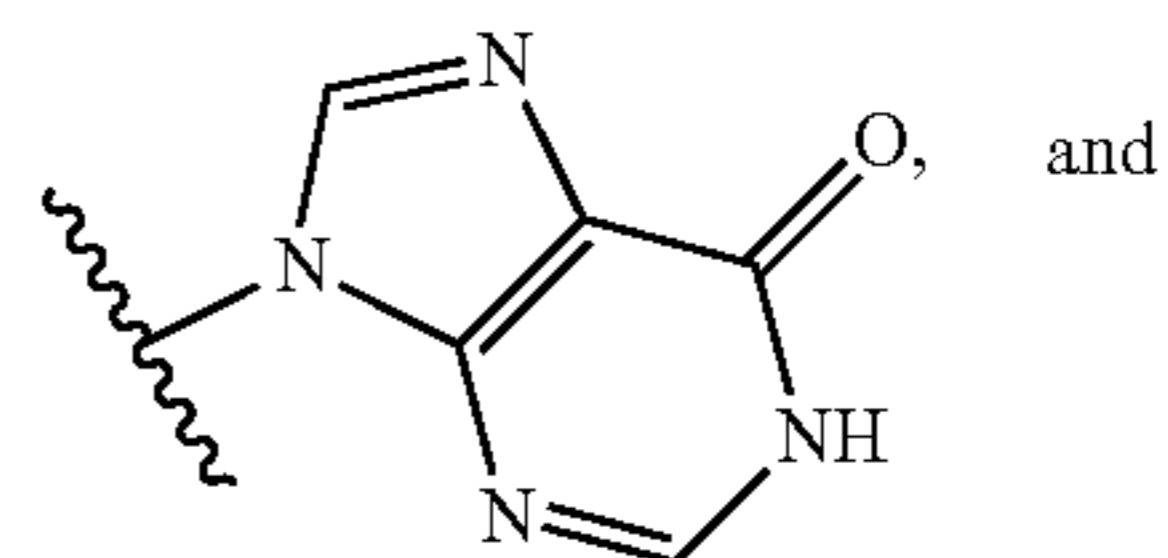
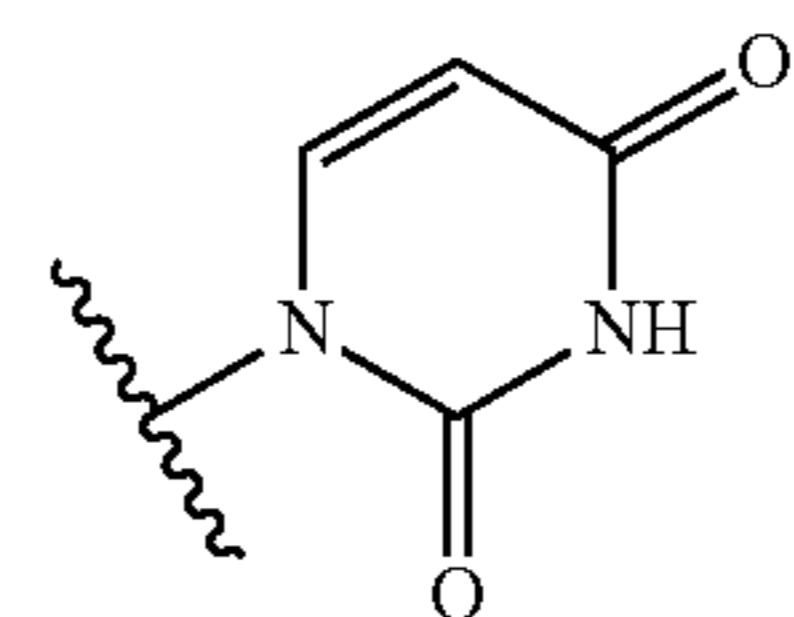


and R⁴ is, independently for each occurrence, selected from the group consisting of:



216

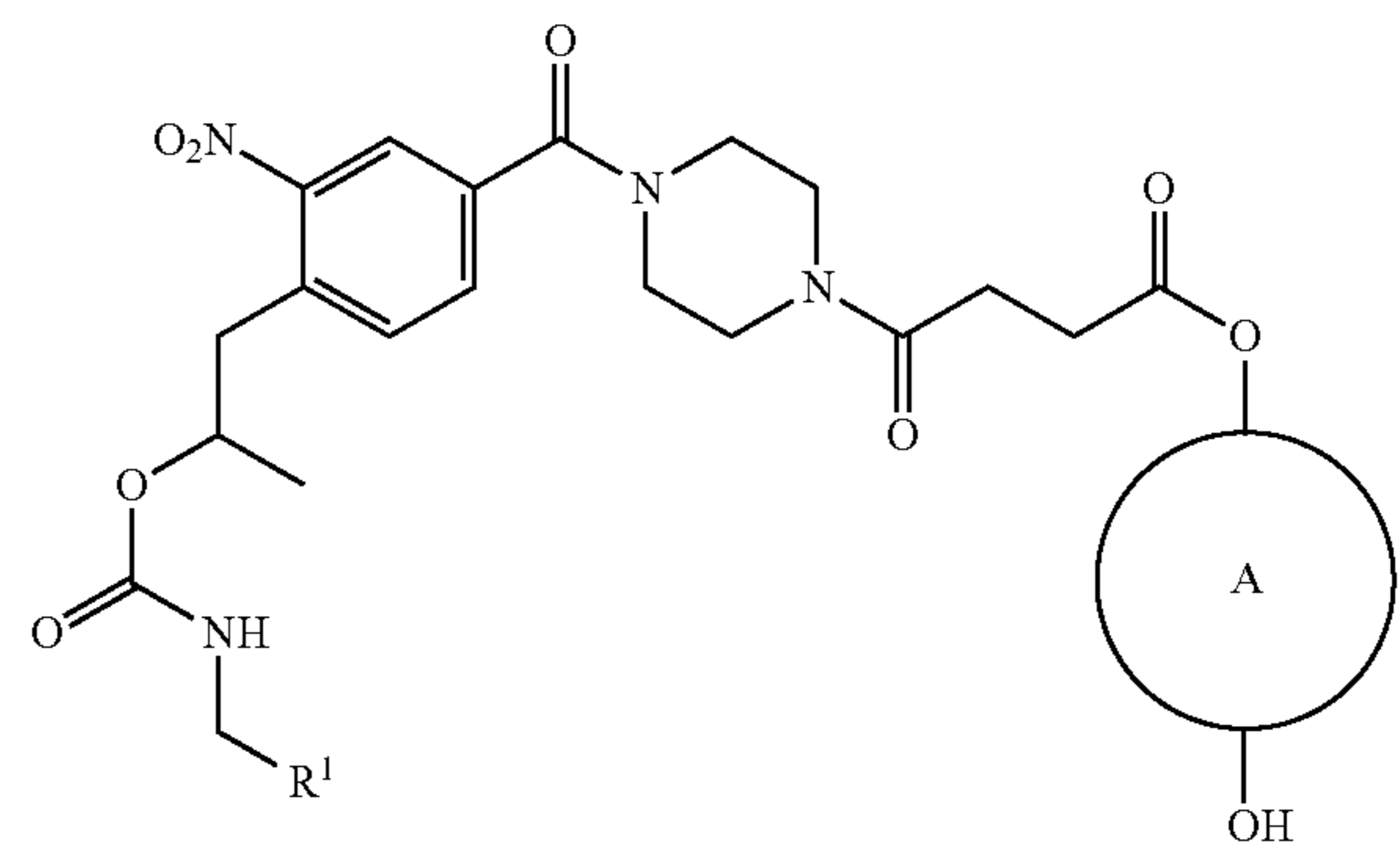
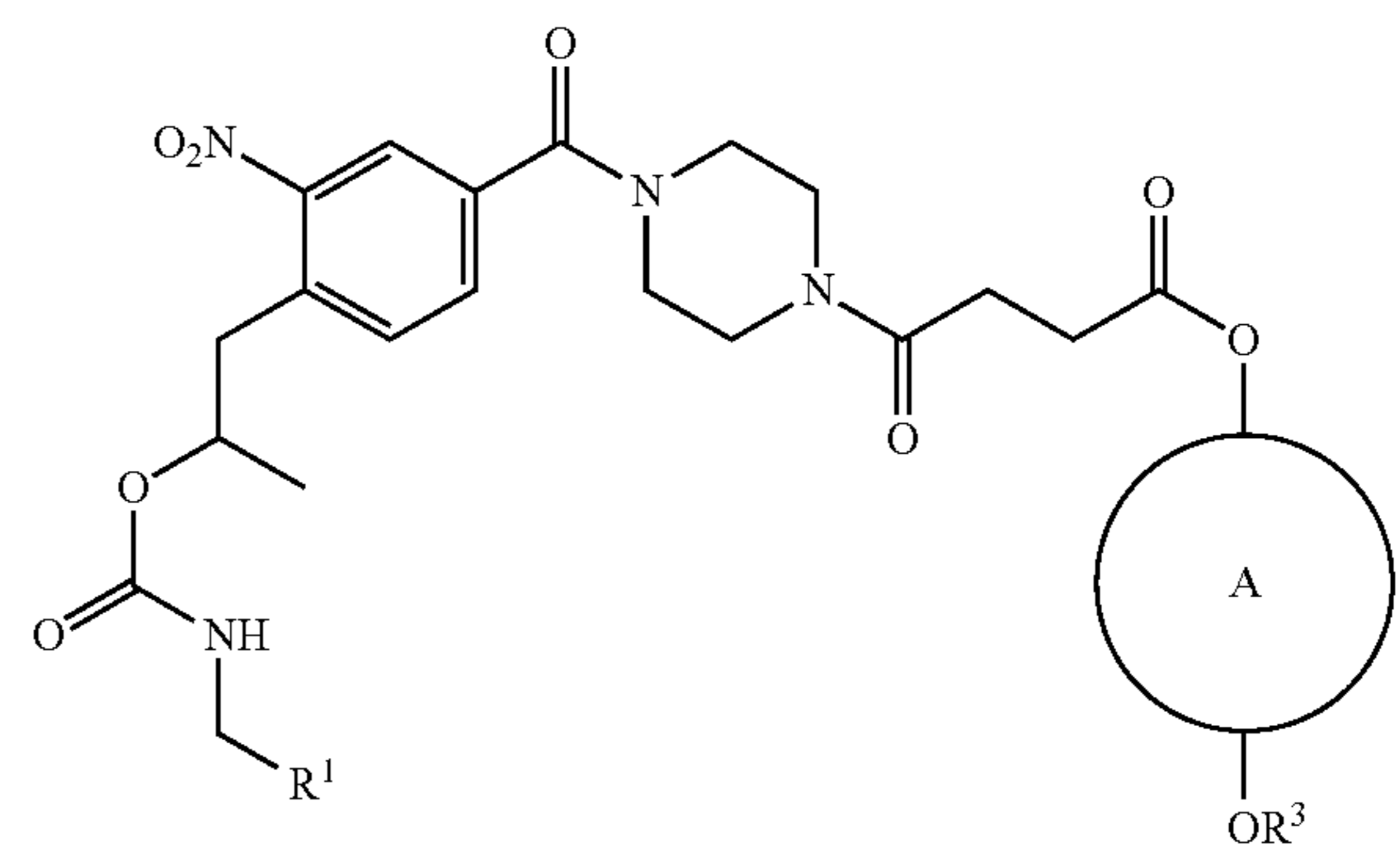
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and

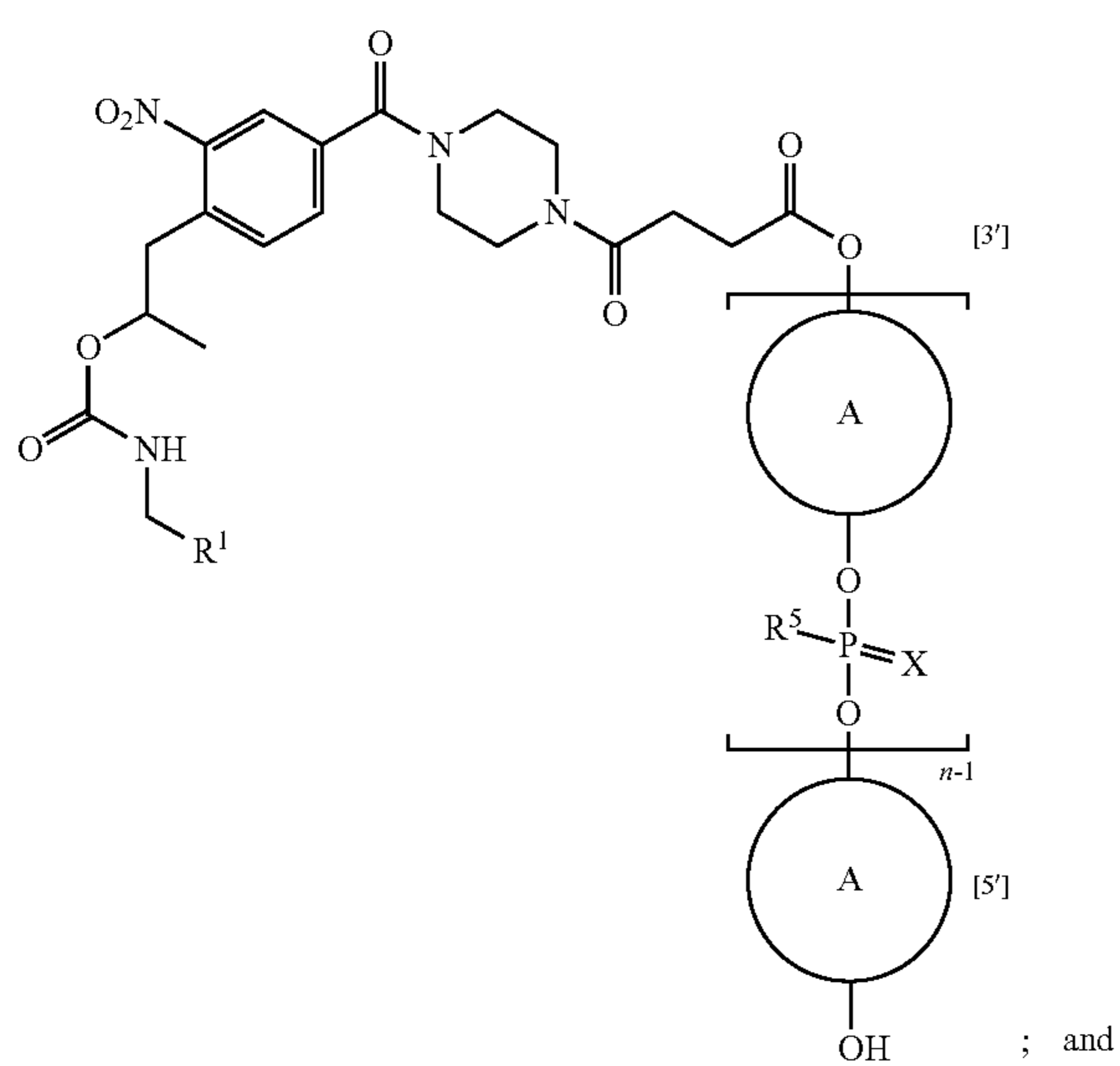
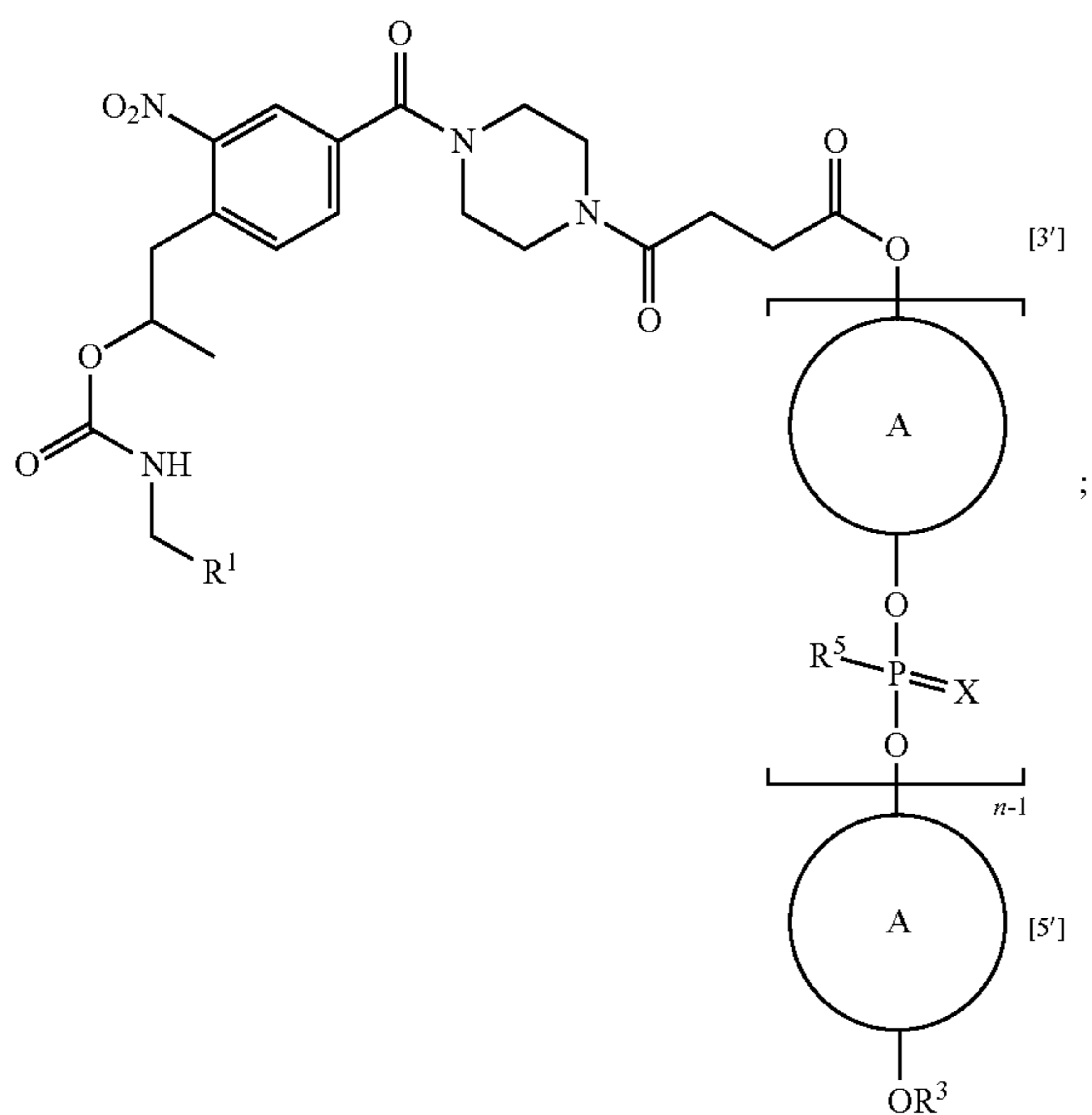
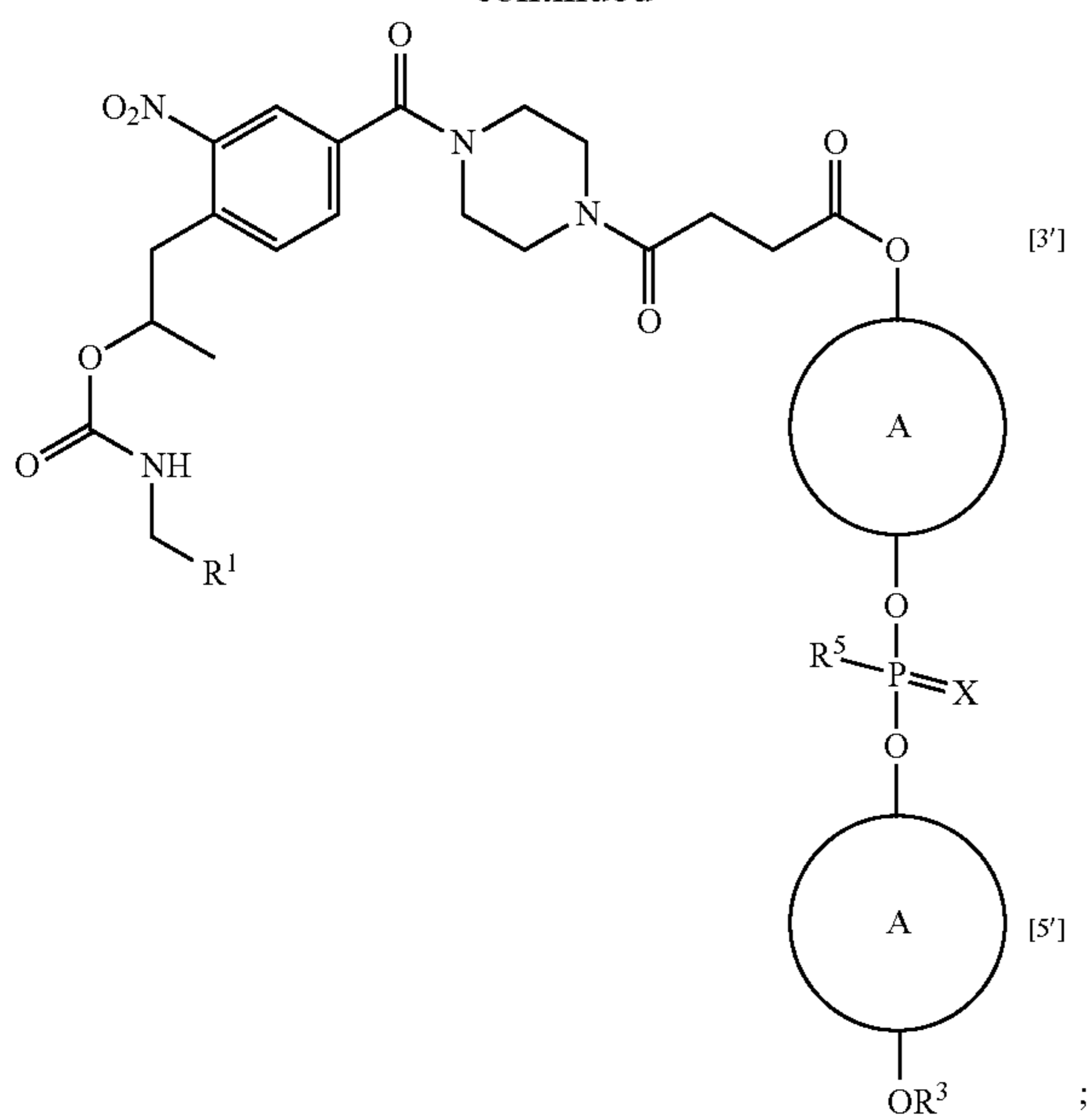
(h) contacting the compound of Formula (F11) with a deprotecting agent to form the oligomeric compound of Formula (F).

The disclosure further provides a compound selected from:



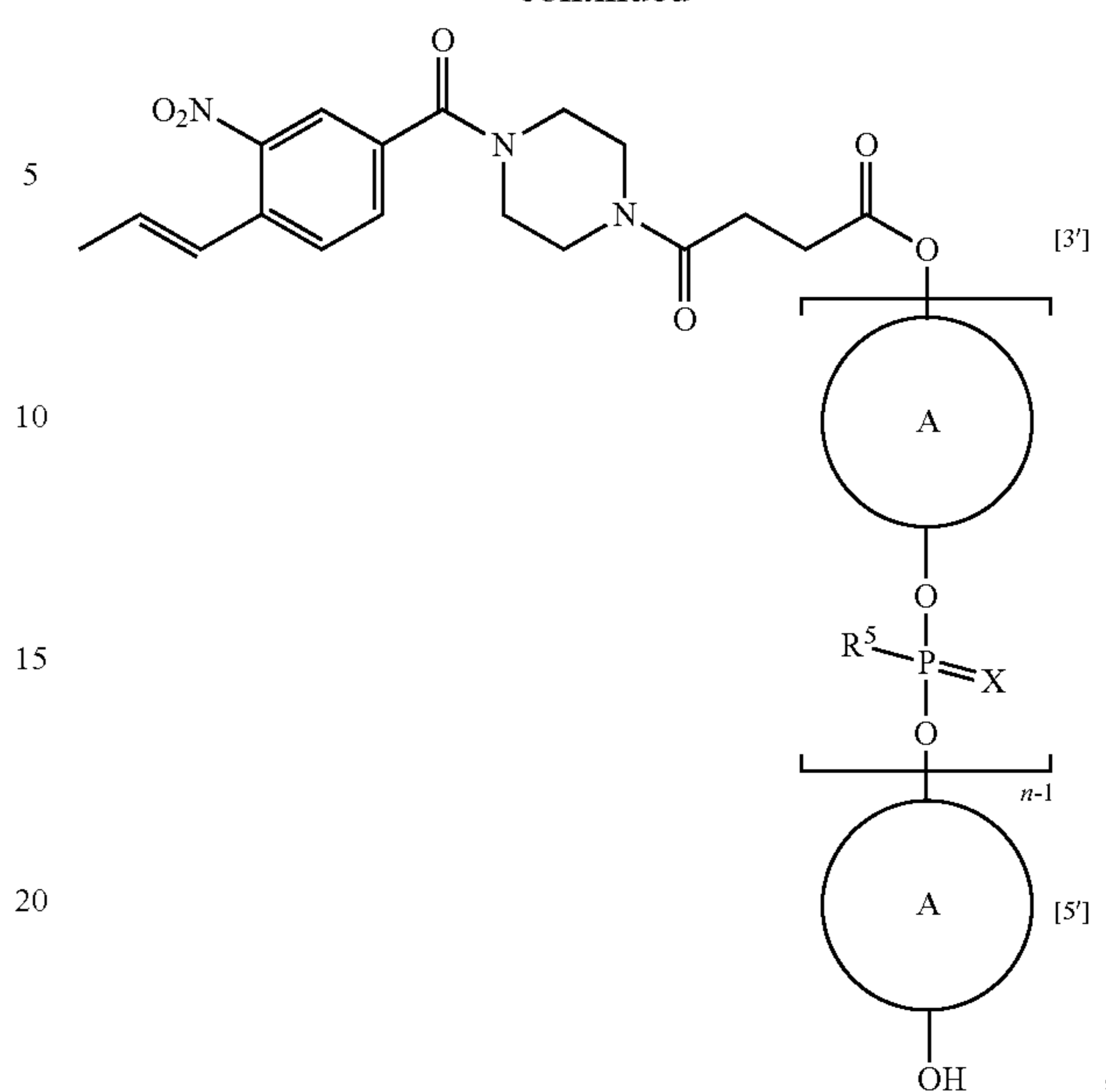
217

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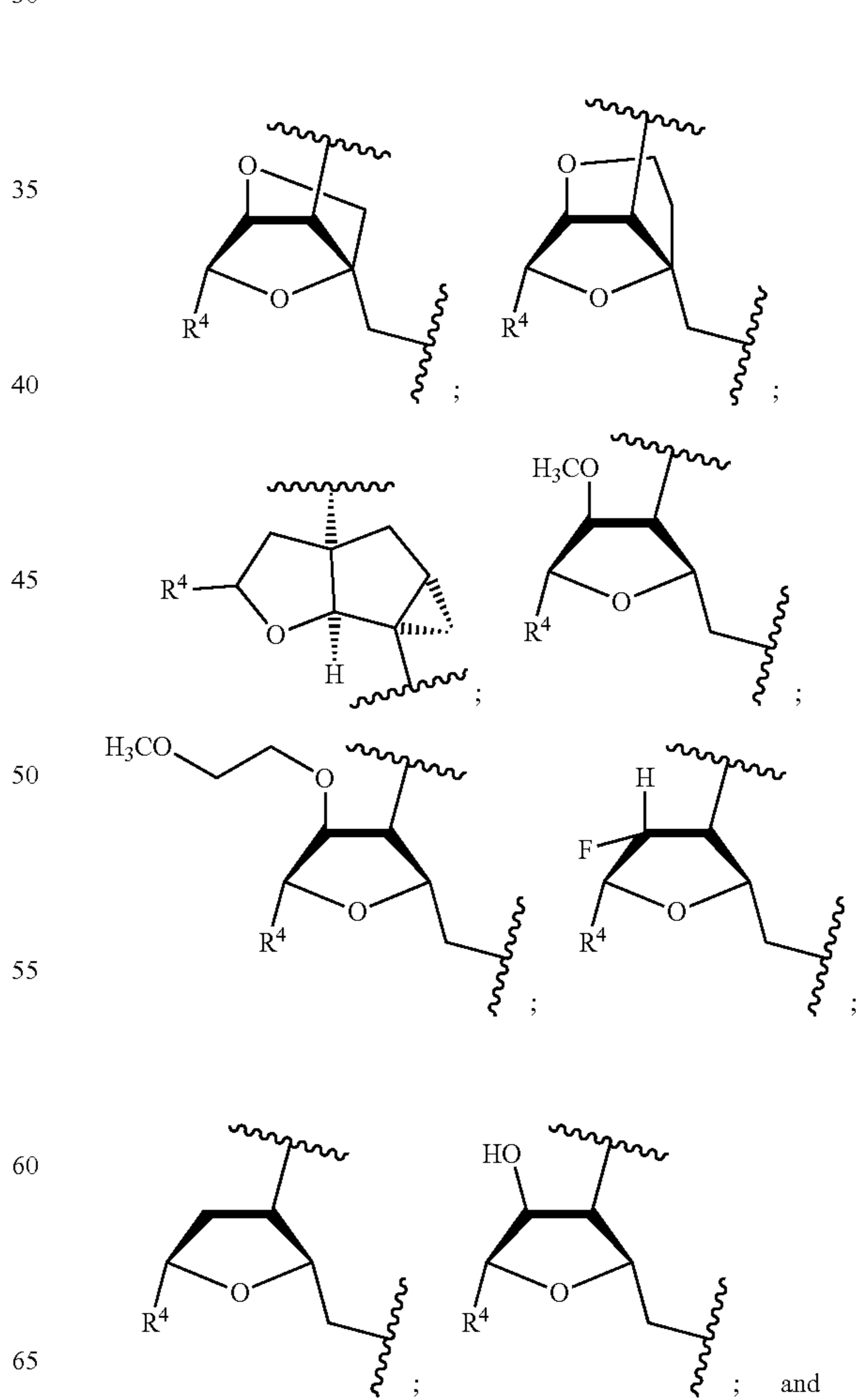
218

-continued



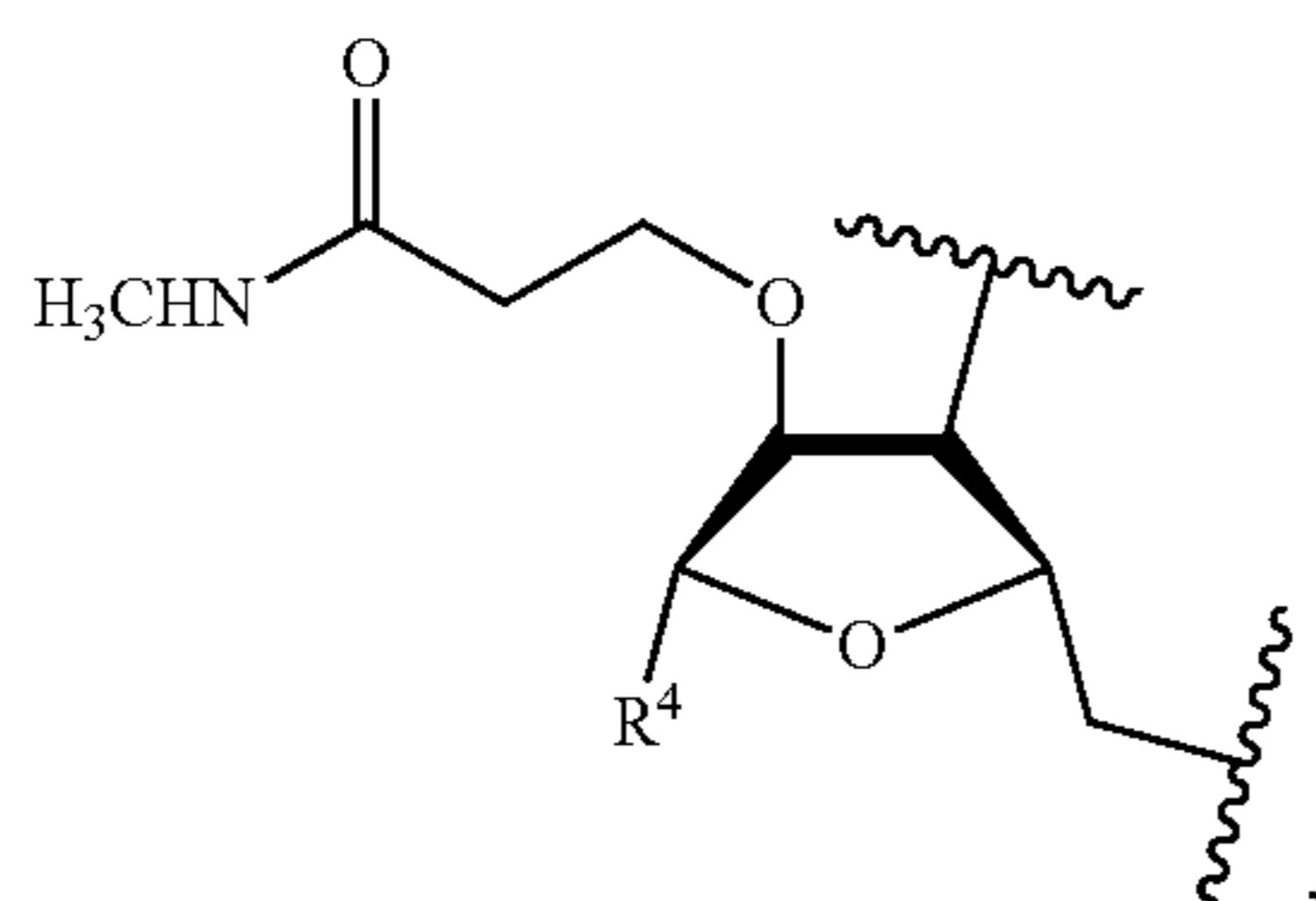
25
30

wherein n is an integer from 10 to 40, R¹ is a support-medium, X is O or S, R⁵ is —OCH₂CH₂CN or —CH₂C(=O)OC(CH₃)₂CH₂CN, each A, independently at each occurrence, is selected from the group consisting of:

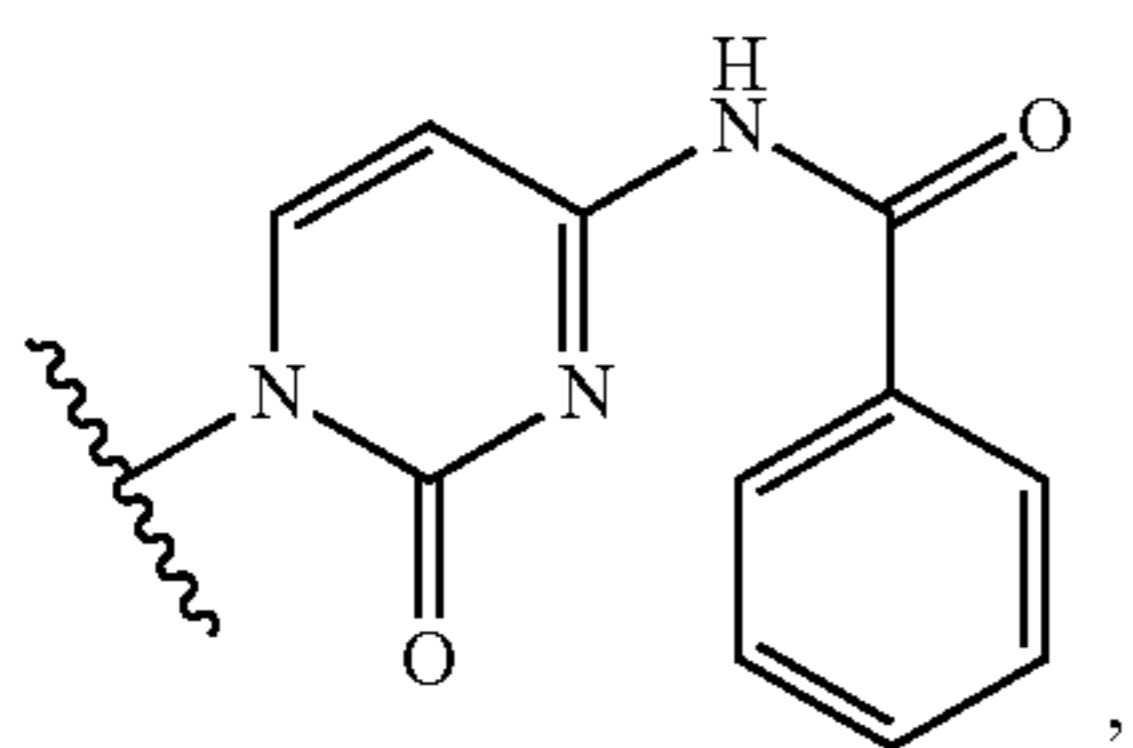


219

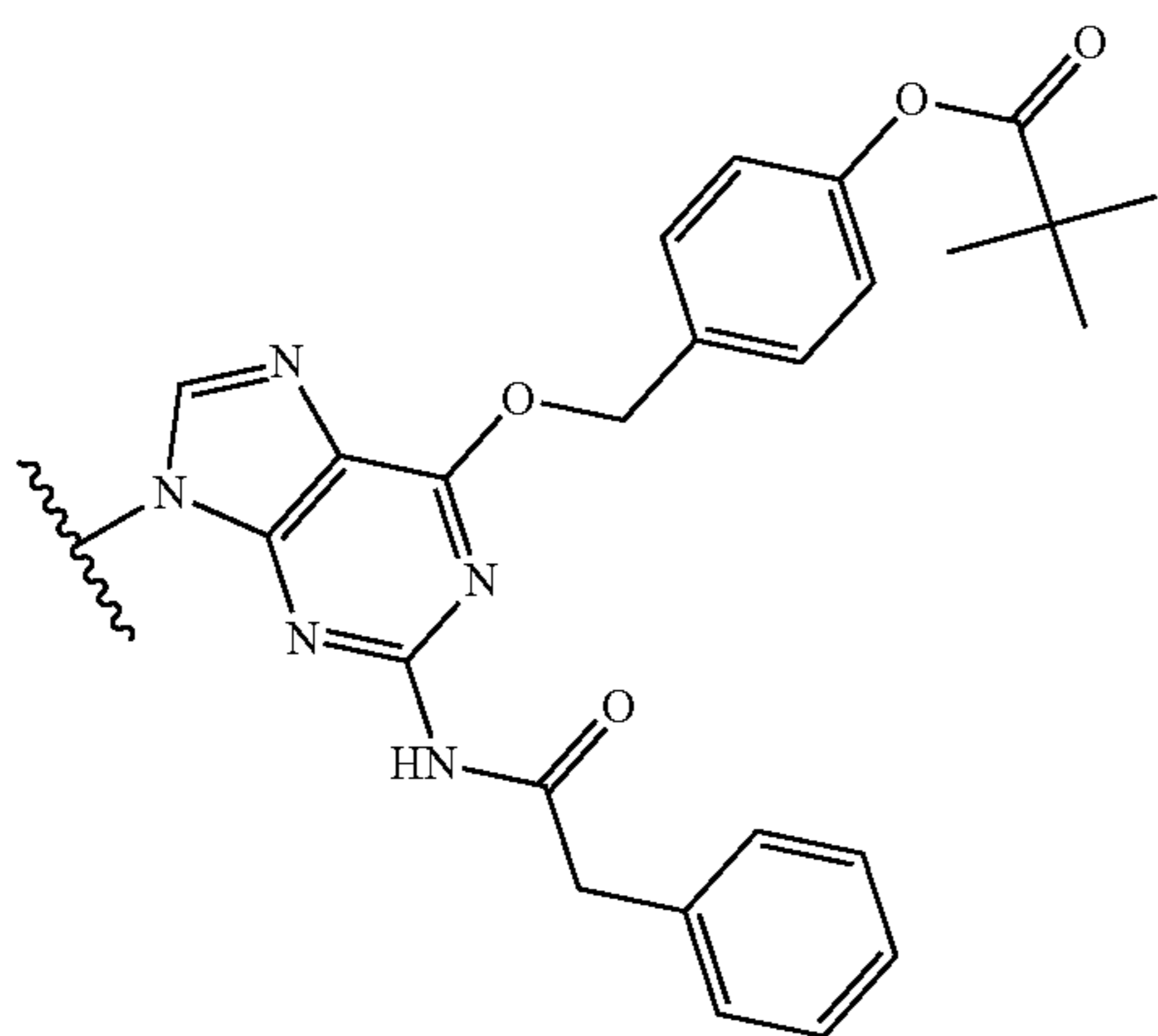
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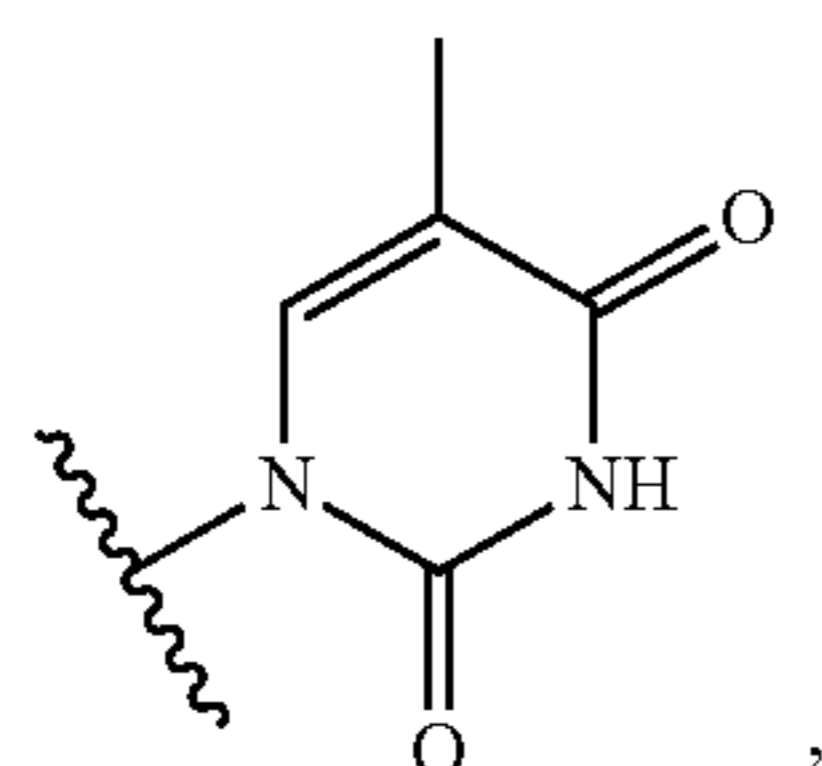
R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and R⁴ is, independently for each occurrence, selected from the group consisting of:



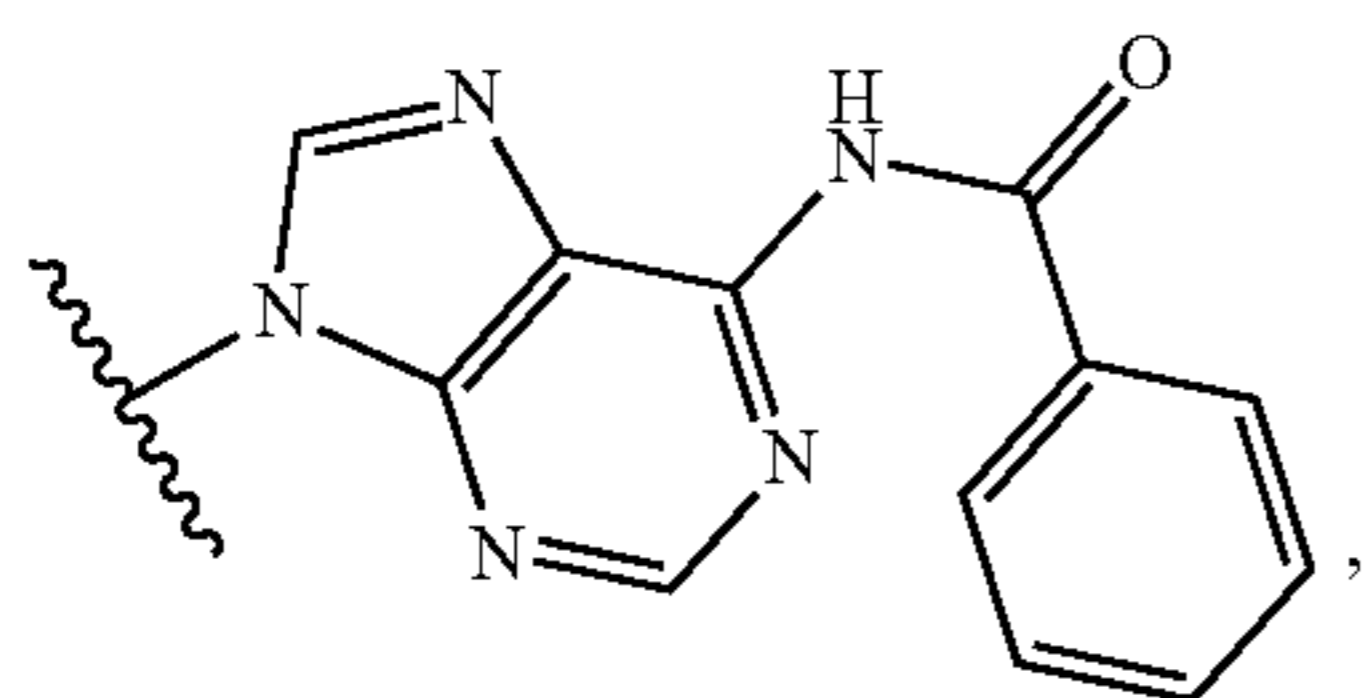
(PC)



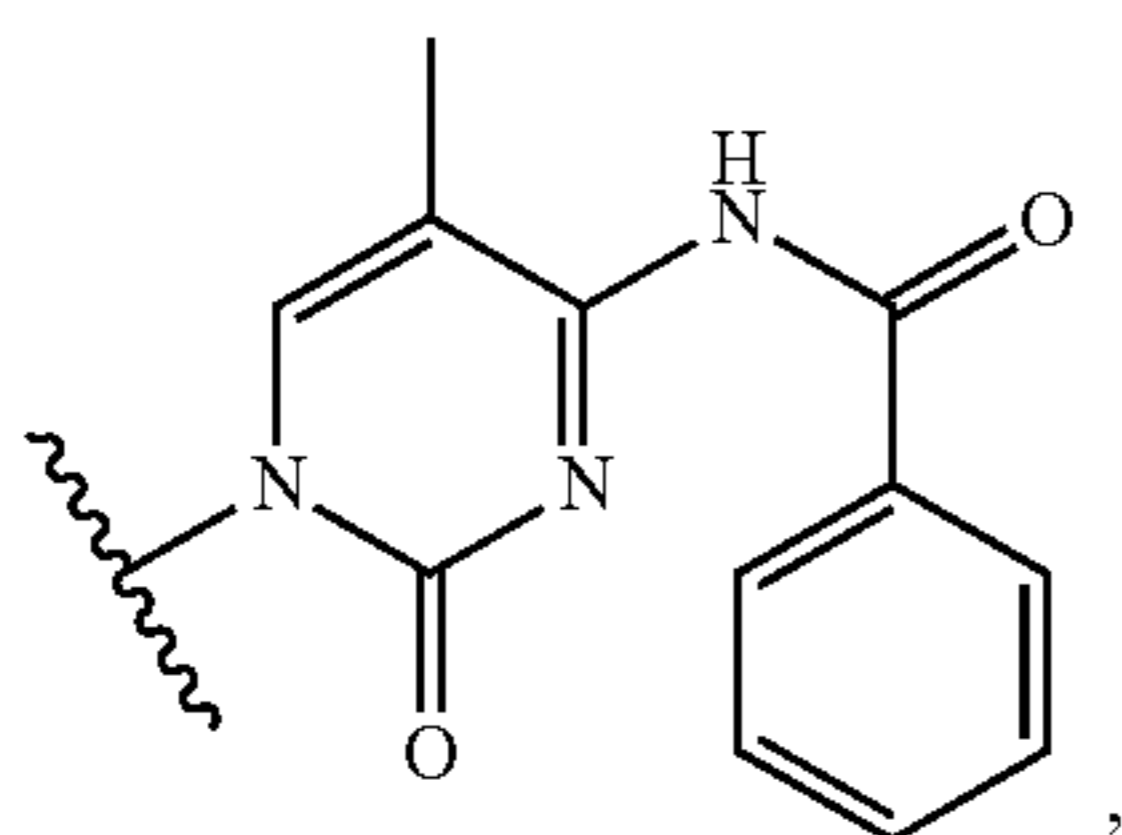
(DPG)



(T)



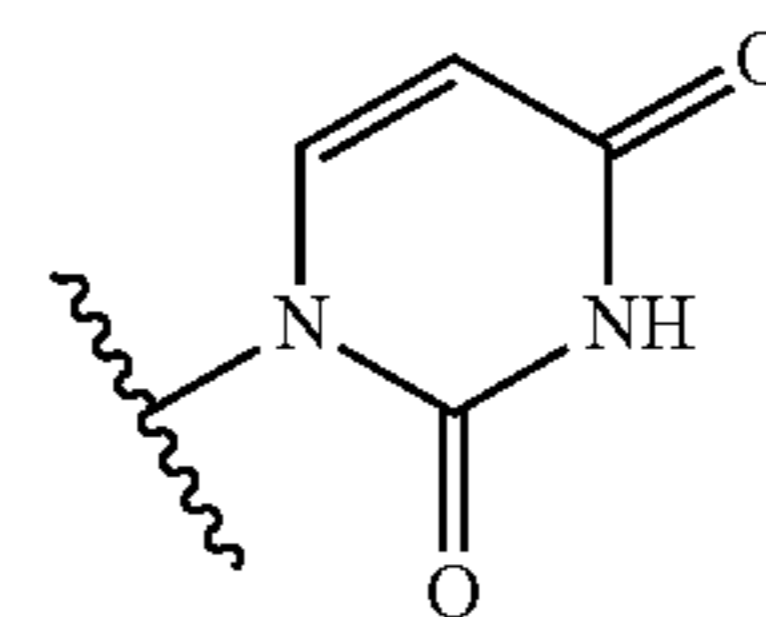
(PA)



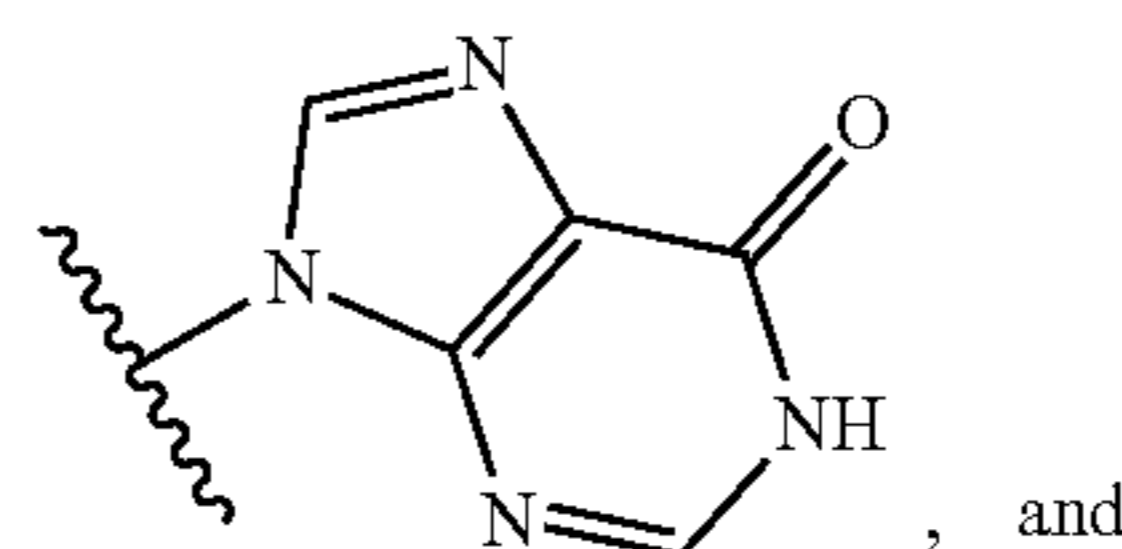
(P5mC)

220

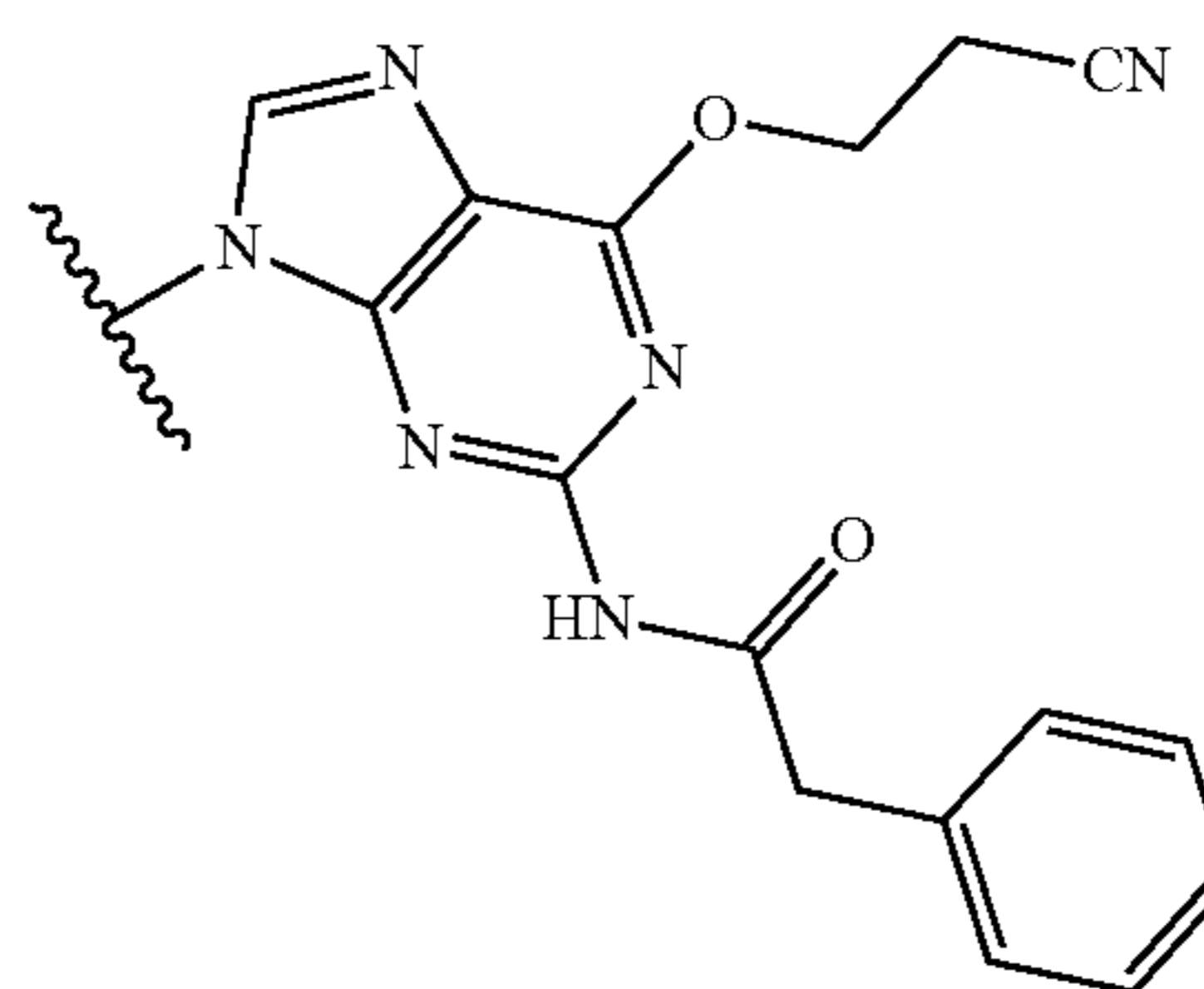
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(U)



(I)



(PG)

Morpholino Oligomers

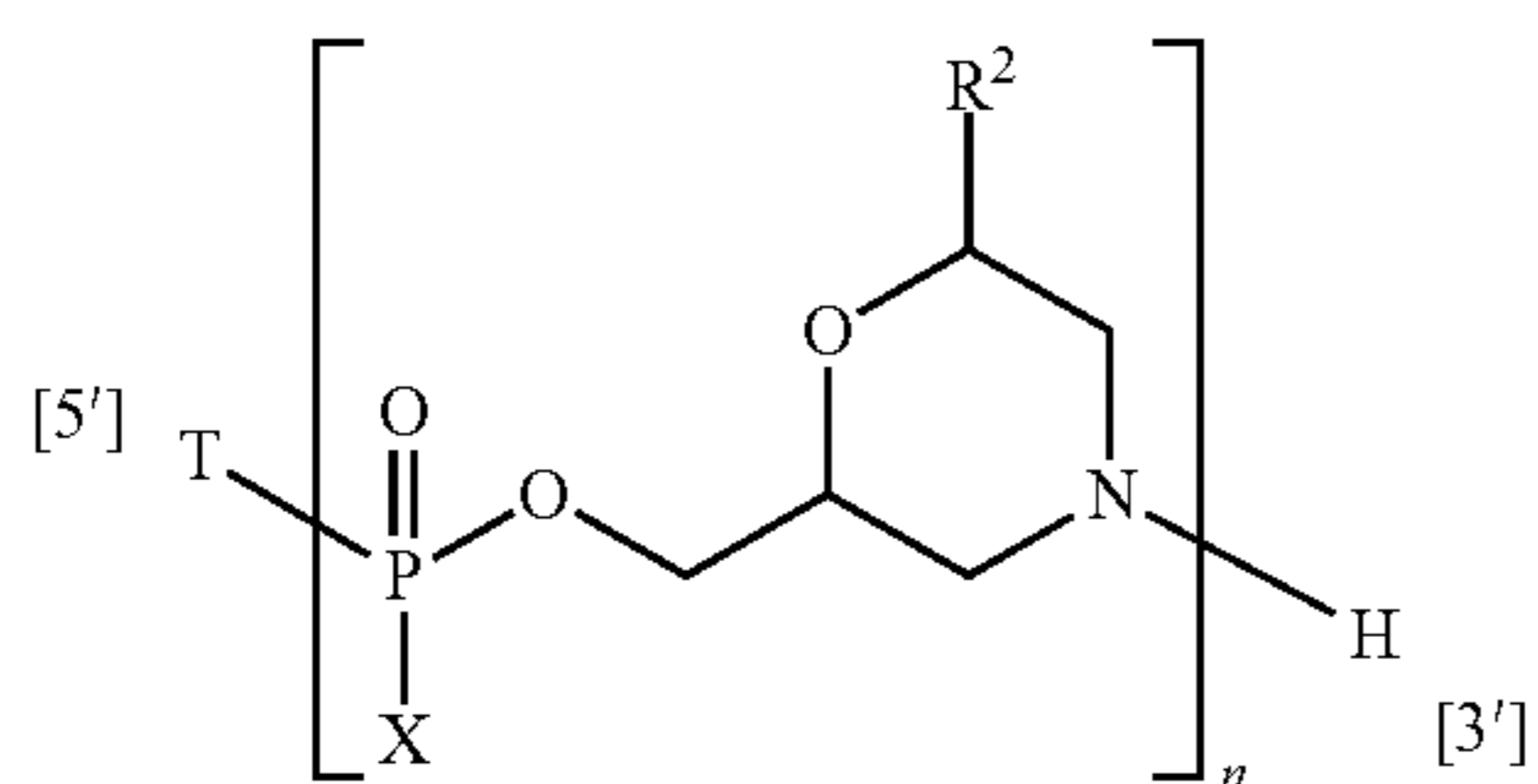
Important properties of morpholino-based subunits include: 1) the ability to be linked in an oligomeric form by stable, uncharged or positively charged backbone linkages; 2) the ability to support a nucleotide base (e.g. adenine, cytosine, guanine, thymidine, uracil, 5-methyl-cytosine and hypoxanthine) such that the polymer formed can hybridize with a complementary-base target nucleic acid, including target RNA; 3) the ability of the oligomer to be actively or passively transported into mammalian cells; and 4) the ability of the oligomer and oligomer:RNA heteroduplex to resist RNase and RNase H degradation, respectively.

In some embodiments, the antisense oligomers contain base modifications or substitutions. For example, certain nucleobases may be selected to increase the binding affinity of the antisense oligomers described herein. 5-methylcytosine substitutions have been shown to increase nucleic acid duplex stability by 0.6-1.2° C., and may be incorporated into the antisense oligomers described herein. In one embodiment, at least one pyrimidine base of the oligomer comprises a 5-substituted pyrimidine base, wherein the pyrimidine base is selected from the group consisting of cytosine, thymine and uracil. In one embodiment, the 5-substituted pyrimidine base is 5-methylcytosine. In another embodiment, at least one purine base of the oligomer comprises hypoxanthine.

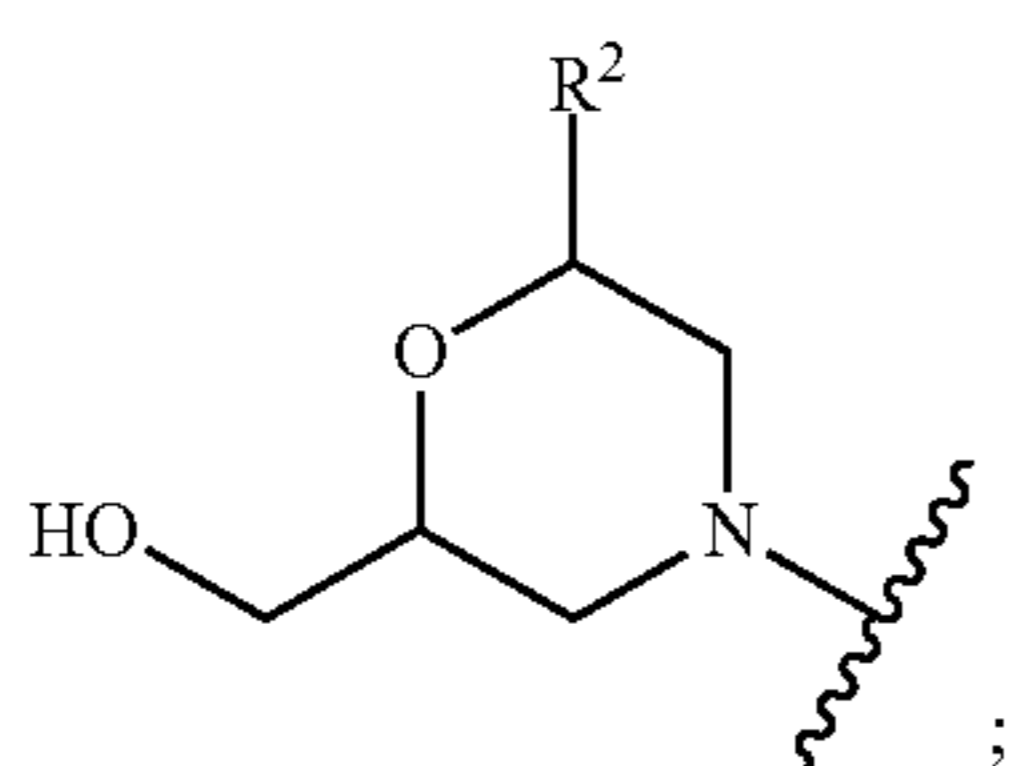
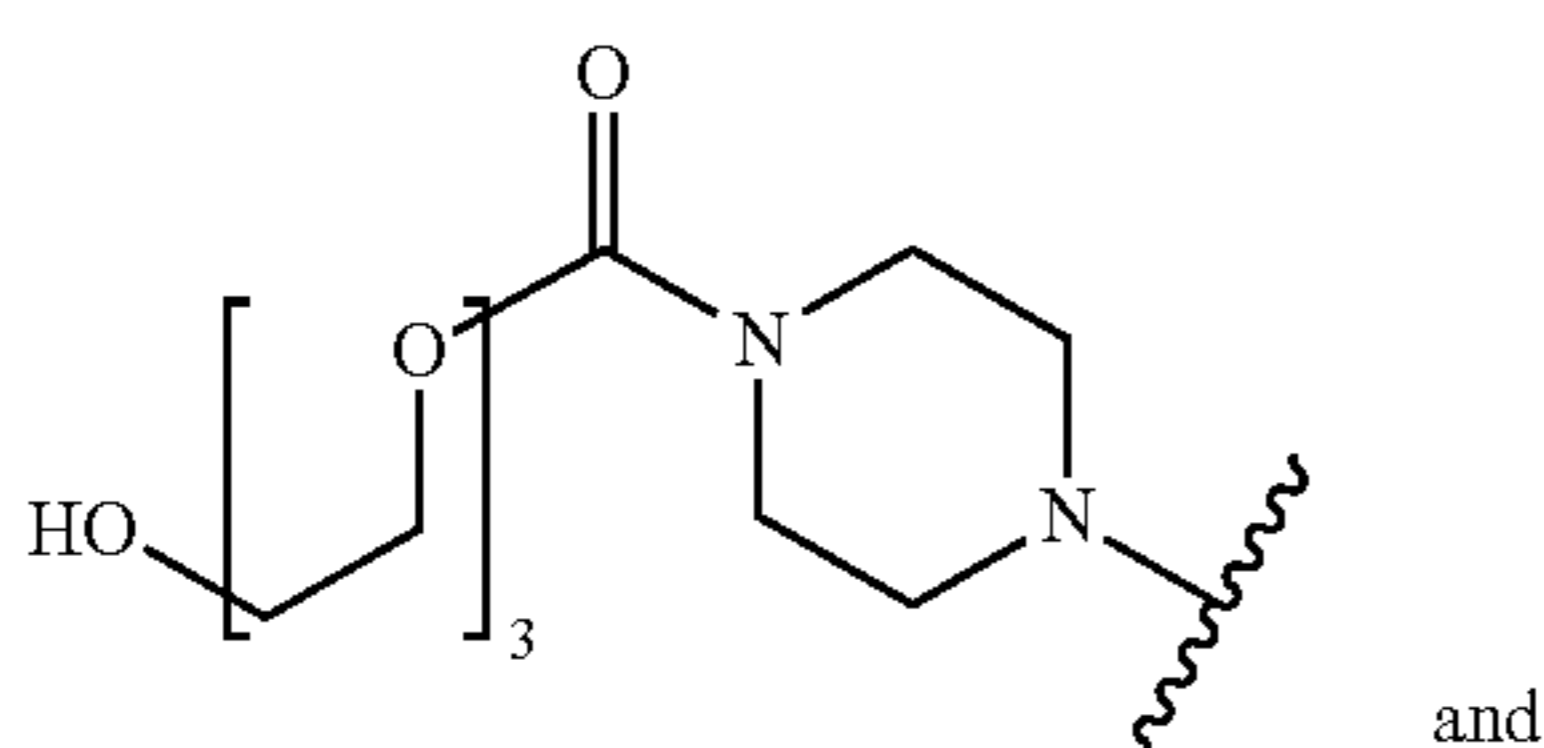
Morpholino-based oligomers (including antisense oligomers) are detailed, for example, in U.S. Pat. Nos. 5,698,685, 5,217,866, 5,142,047, 5,034,506, 5,166,315, 5,185,444, 5,521,063, 5,506,337, 8,299,206, and 8,076,476; International Patent Application Publication Nos. WO/2009/064471 and WO/2012/043730; and Summerton et al., *Anti-sense Nucleic Acid Drug Dev.* 1997, 7, 187-195, each of which are hereby incorporated by reference in their entirety.

In some embodiments, the disclosure provides a process for preparing an oligomeric compound of Formula (B).

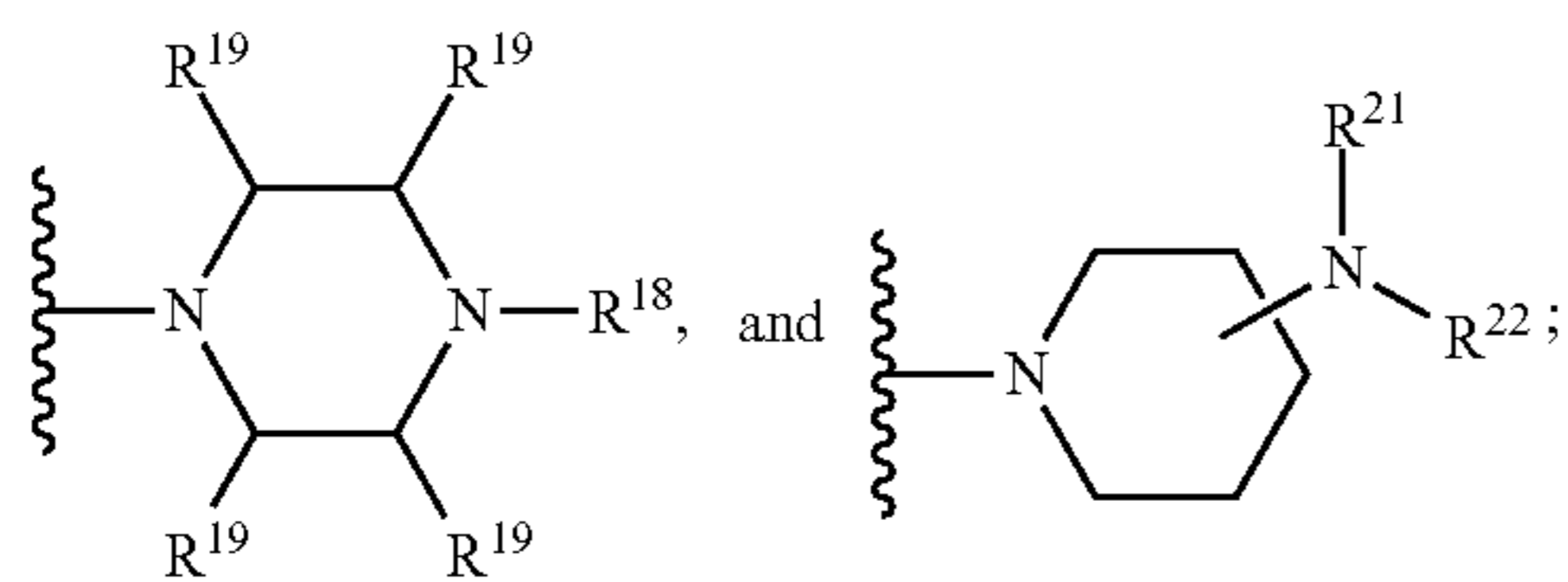
221



wherein n is an integer from 10 to 40,
T is selected from



X is, independently at each occurrence, selected from H,
OH, —OCH₂CH₂CN, N(R¹⁷)₂,



wherein

R¹⁷ is, independently for each occurrence, H or C₁₋₆-alkyl;

R¹⁸ is selected from the group consisting of H, C₁₋₆-alkyl,
—C(=NH)NH₂, —C(O)(CH₂)_qNR²⁰C(=NH)NH₂,
and —C(O)(CH₂)₂NHC(O)(CH₂)₅NR²⁰C(=NH)NH₂;

q is 1, 2, 3, 4, or 5;

R¹⁹ is, independently for each occurrence, H or methyl;

R²⁰ is H or C₁₋₆-alkyl;

R²¹ is selected from the group consisting of H, C₁₋₆-alkyl,
C(=NH)NH₂, C(O)(CH₂)_rNR²³C(=NH)NH₂, C(O)
CH(NH₂)(CH₂)₃NHC(=NH)NH₂, C(O)(CH₂)₂NHC
(O)(CH₂)₅NR²³C(=NH)NH₂, and C(O)CH(NH₂)
(CH₂)₄NH₂;

R²² is H or C₁₋₆-alkyl;

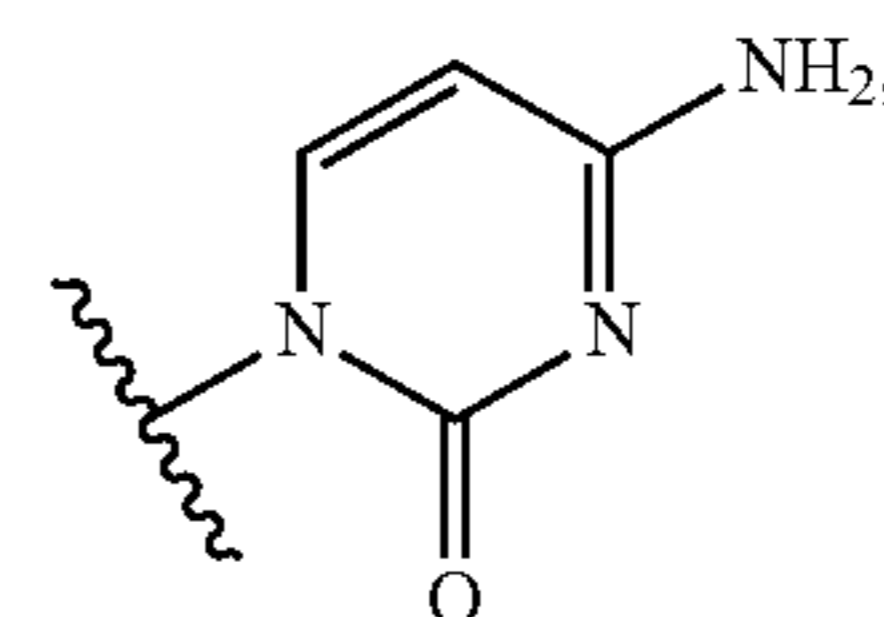
R²³ is H or C₁₋₆-alkyl; and

r is 1, 2, 3, 4, or 5;

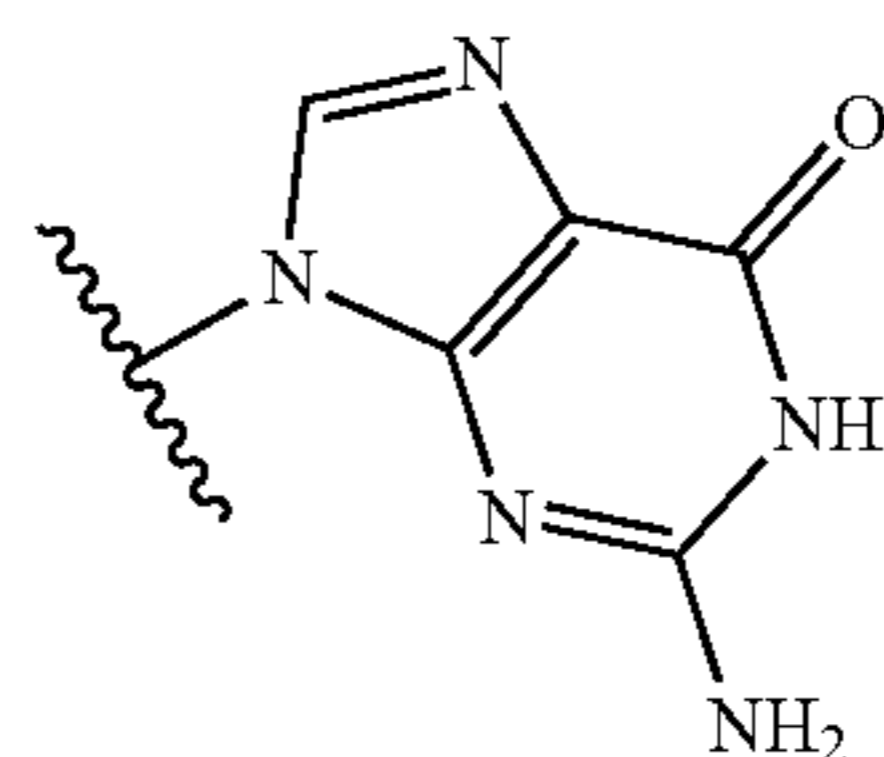
222

(B) each R² is, independently for each occurrence, selected from the group consisting of:

5

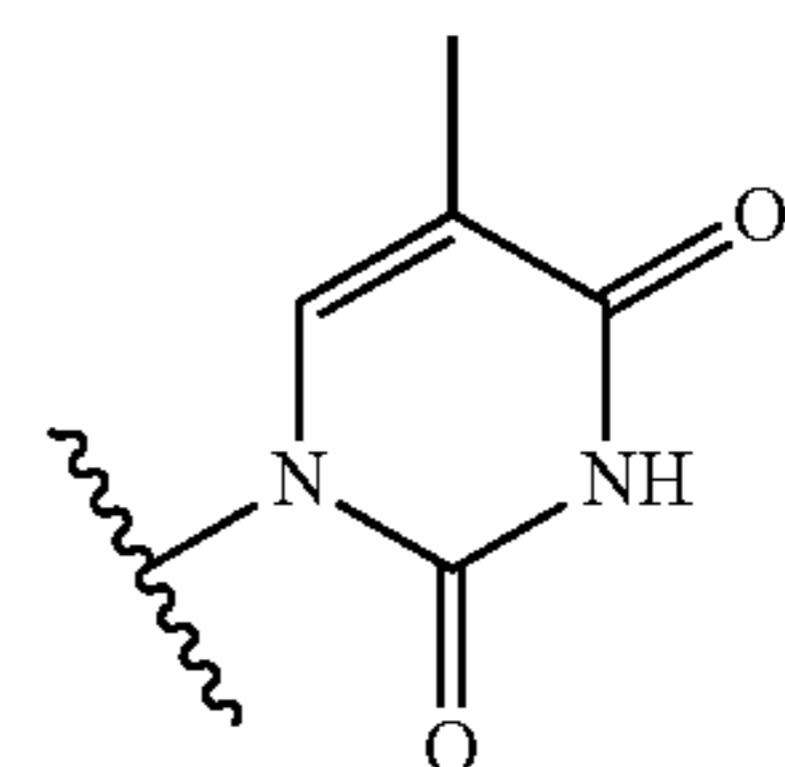


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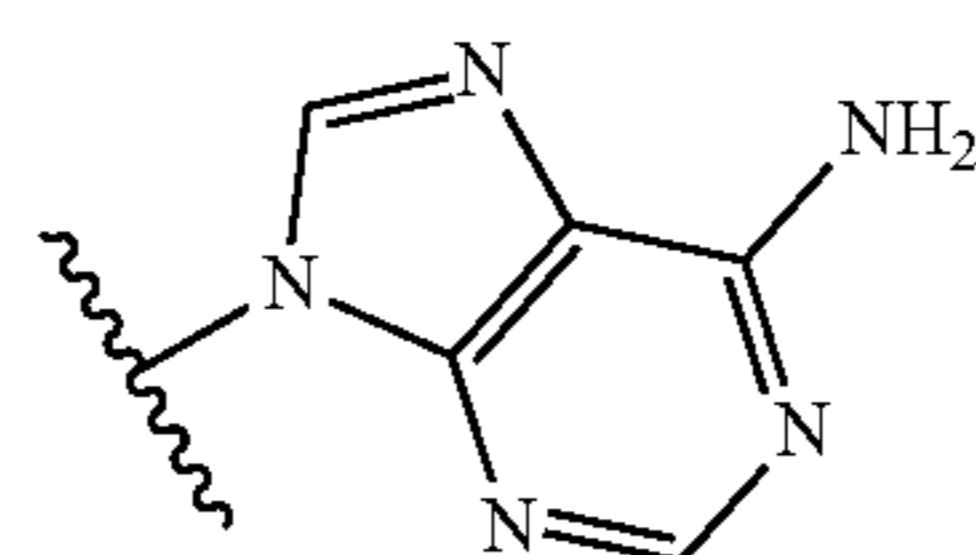
15

20



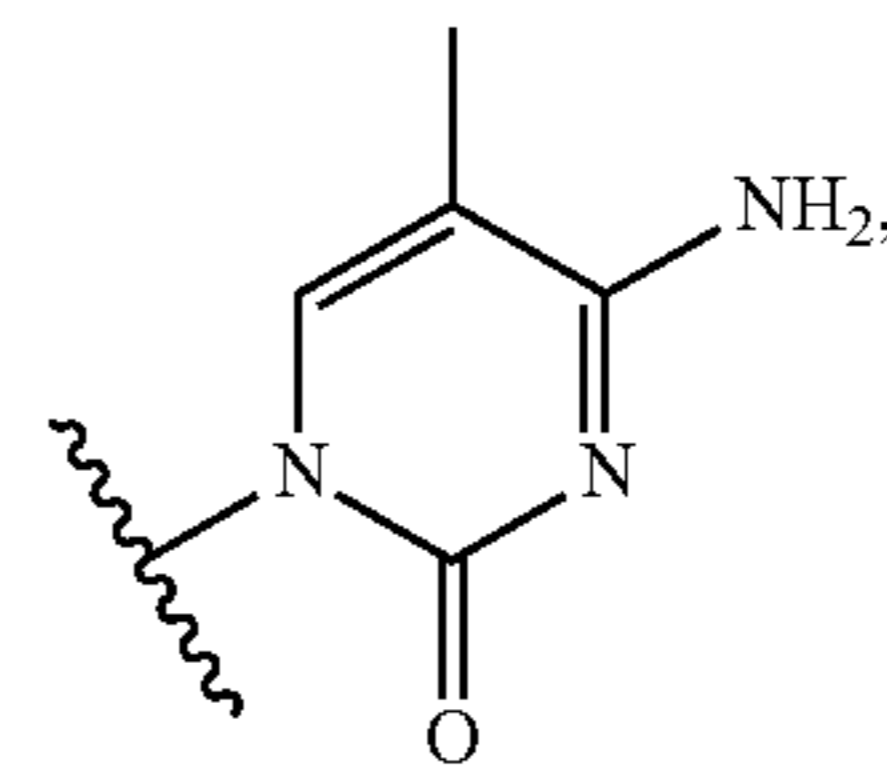
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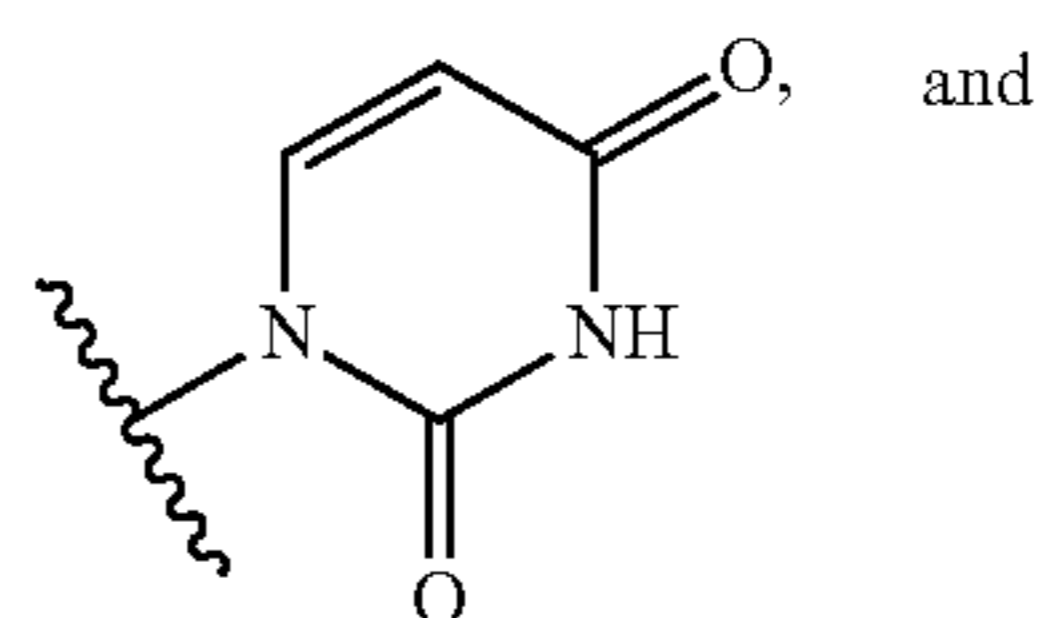
35

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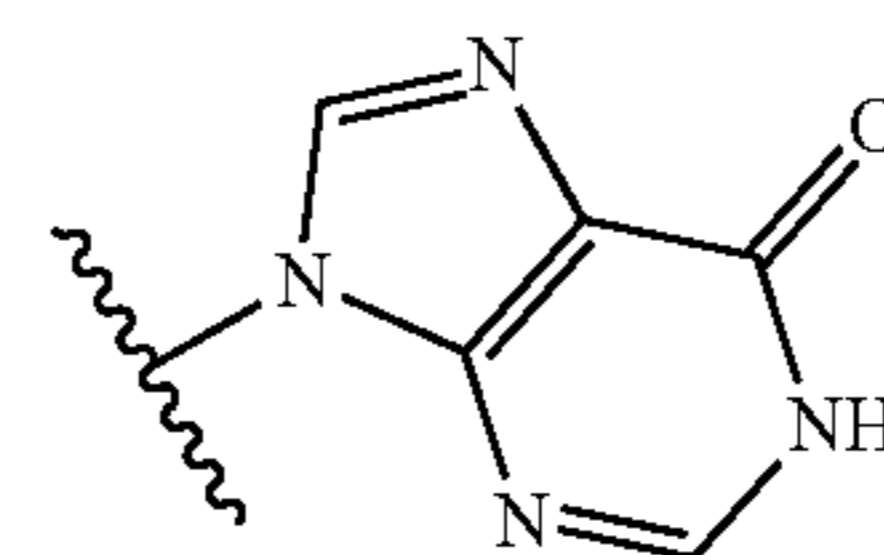
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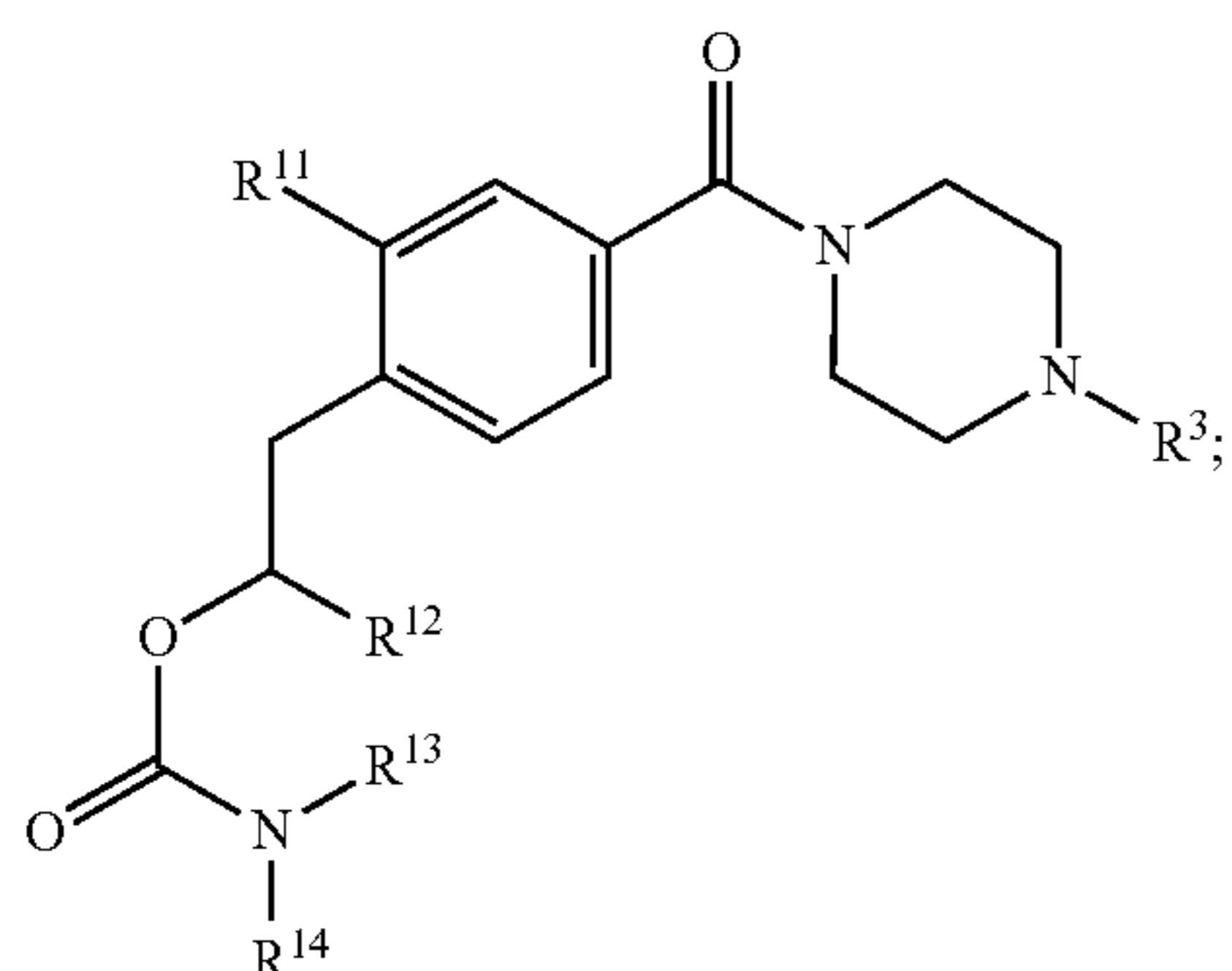
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65



223

wherein the process comprises the sequential steps of:
 (a) contacting a compound of Formula (B1):



wherein

R^{11} is selected from the group consisting of halo, CN, and NO_2 ;

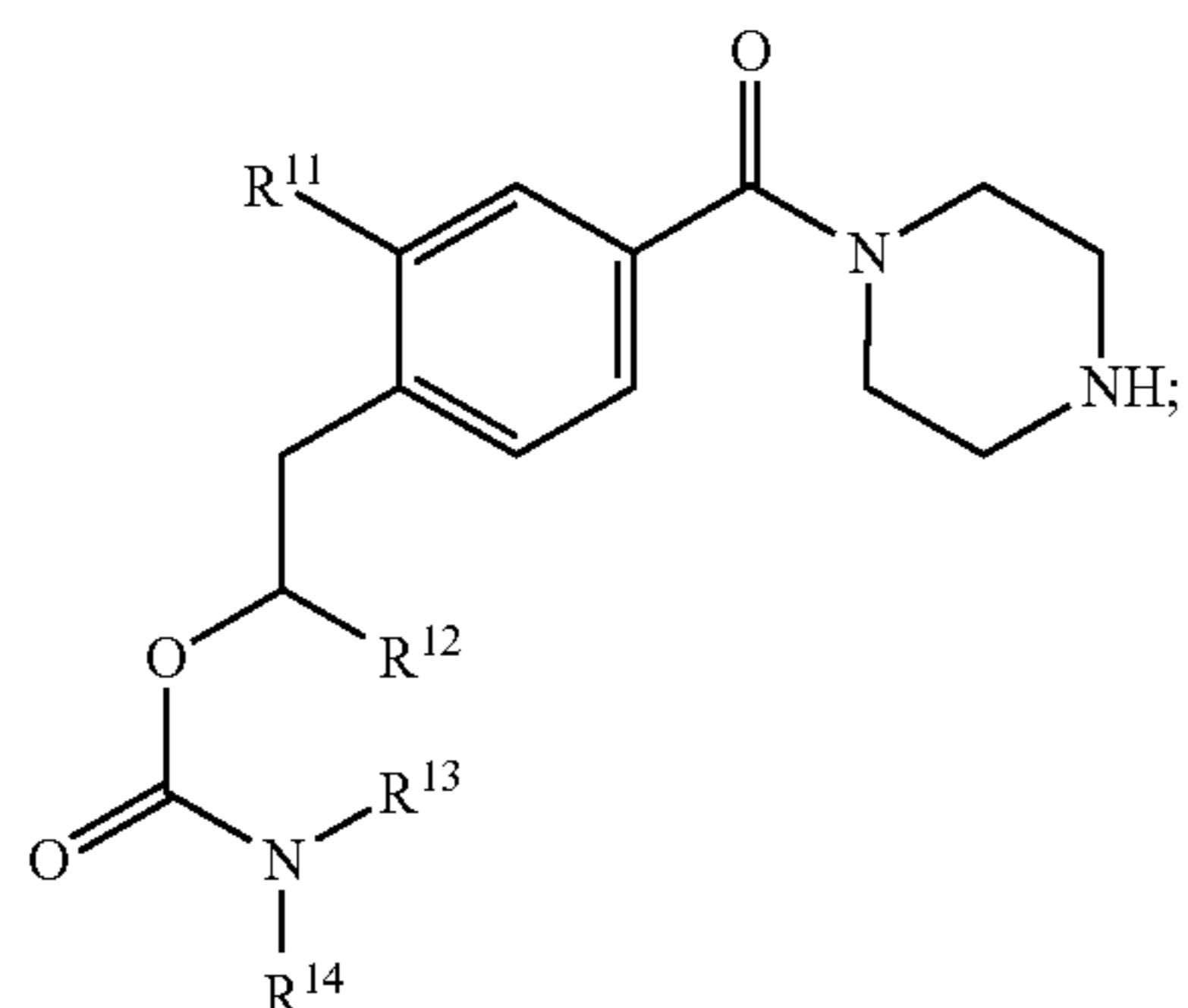
R^{12} is C_{1-6} -alkyl;

R^{13} is H or C_{1-6} -alkyl; and

R^{14} is a first linker bound to a support-medium,

R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

with a deblocking agent to form the compound of Formula (II-B):



wherein

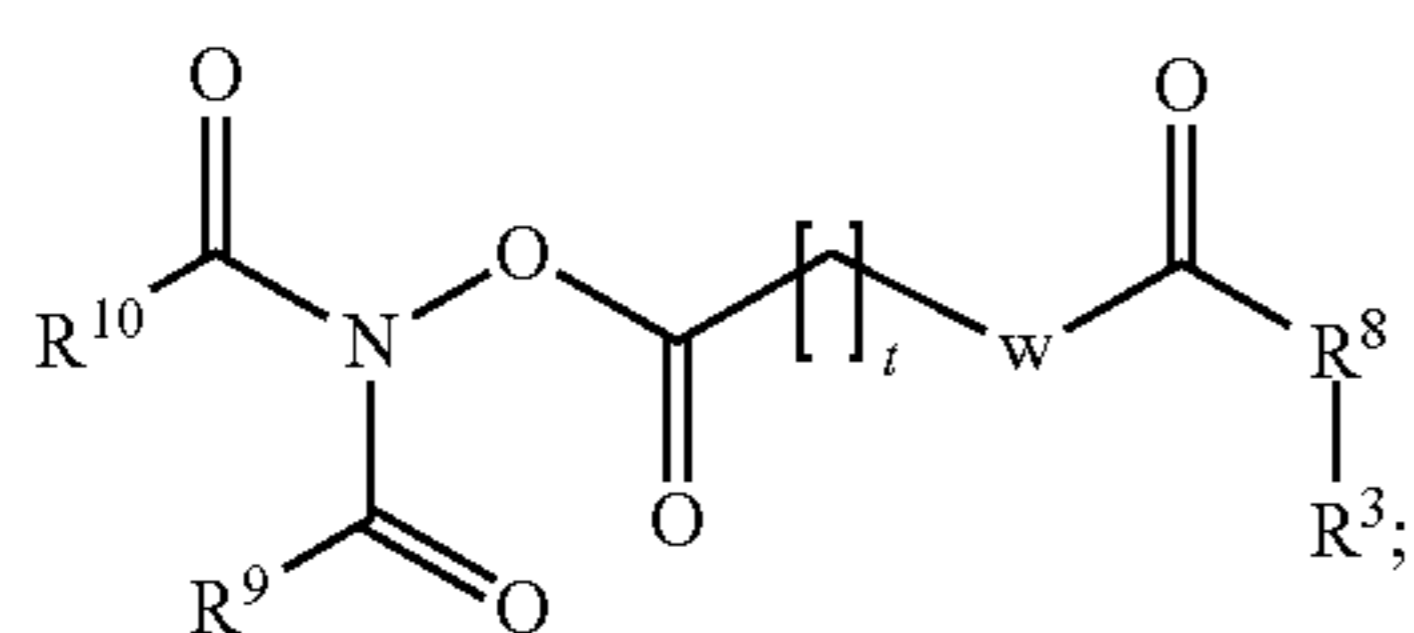
R^{11} is selected from the group consisting of halo, CN, and NO_2 ;

R^{12} is C_{1-6} -alkyl;

R^{13} is H or C_{1-6} -alkyl; and

R^{14} is a first linker bound to a support-medium;

(b) contacting the compound of Formula (II) with a compound of Formula (III):



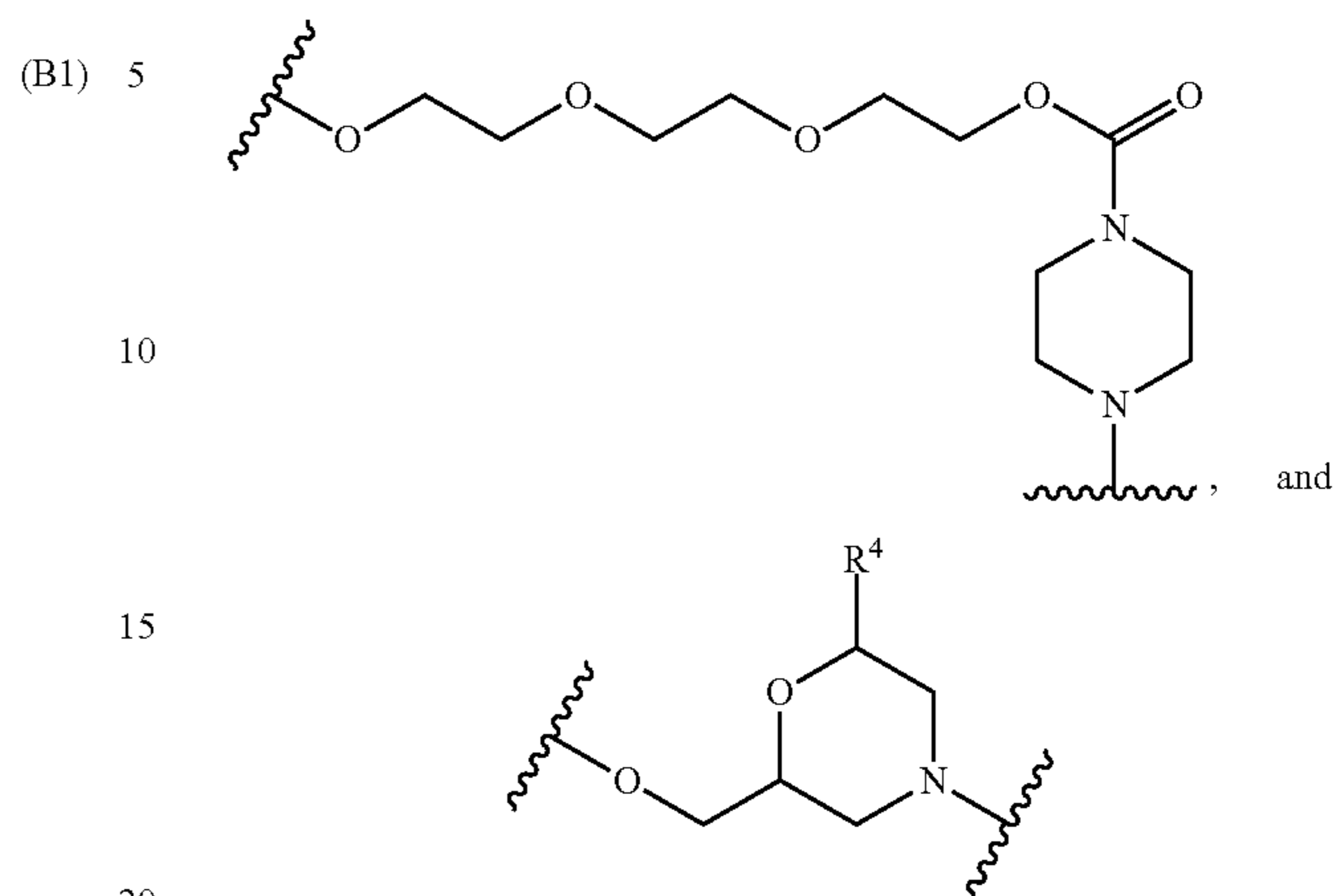
wherein

w is selected from O or C;

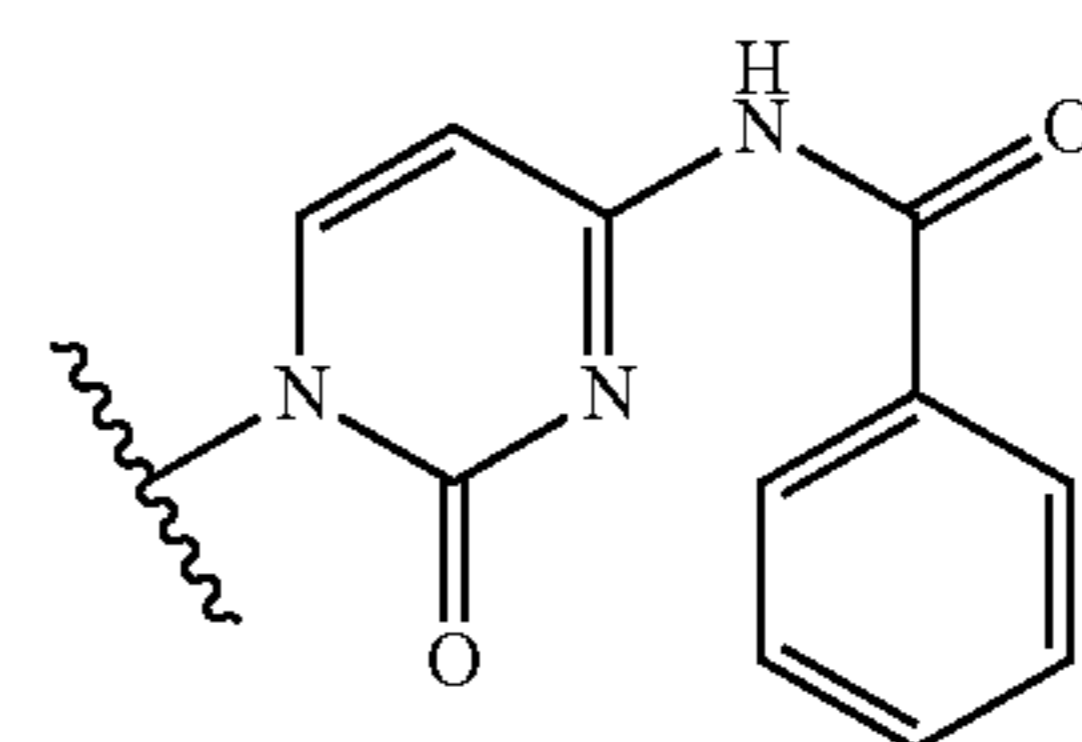
t is 1, 2, 3, 4, or 5;

224

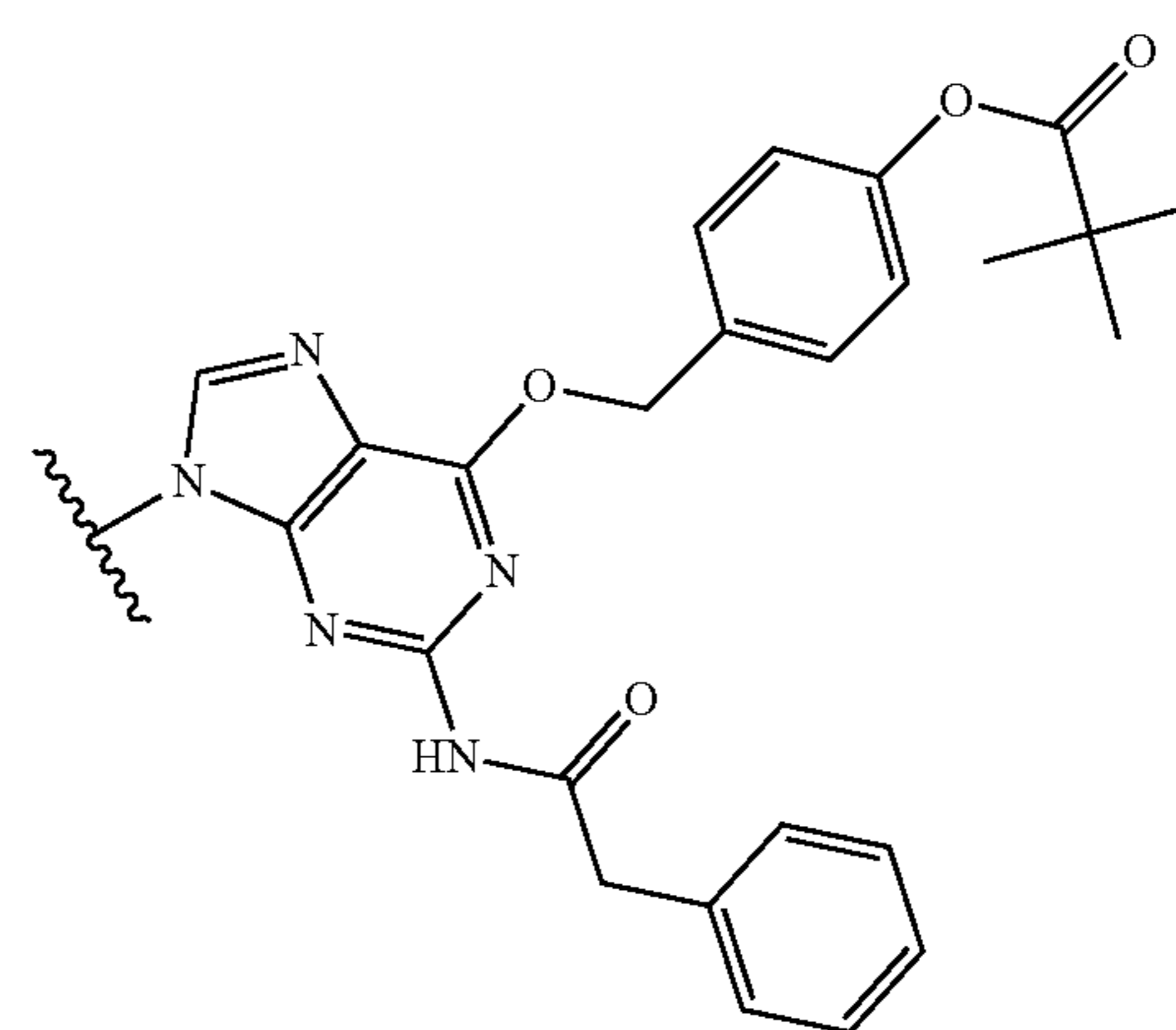
R^8 is selected from:



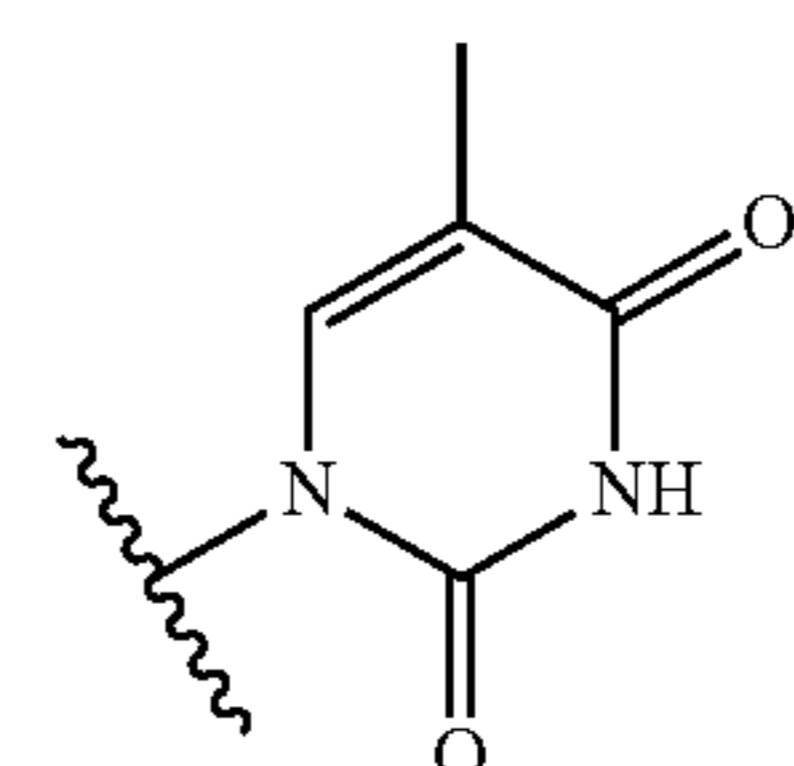
wherein R^4 is selected from the group consisting of:



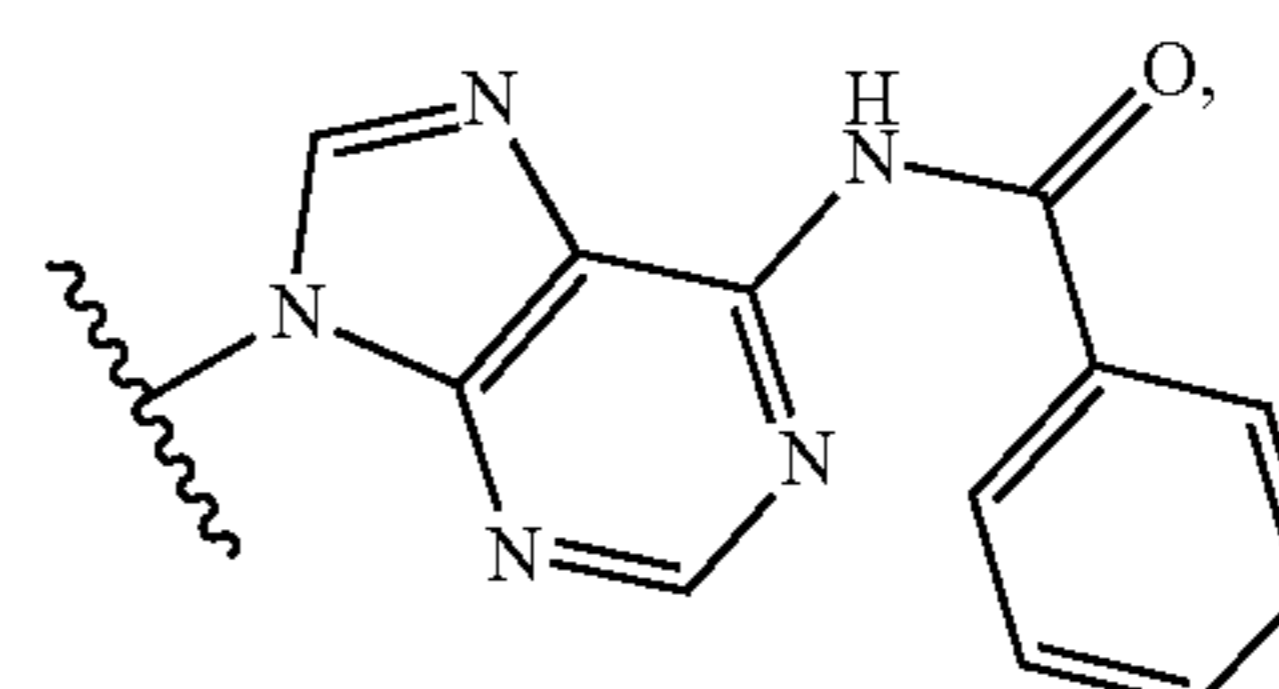
(PC)



(DPG)



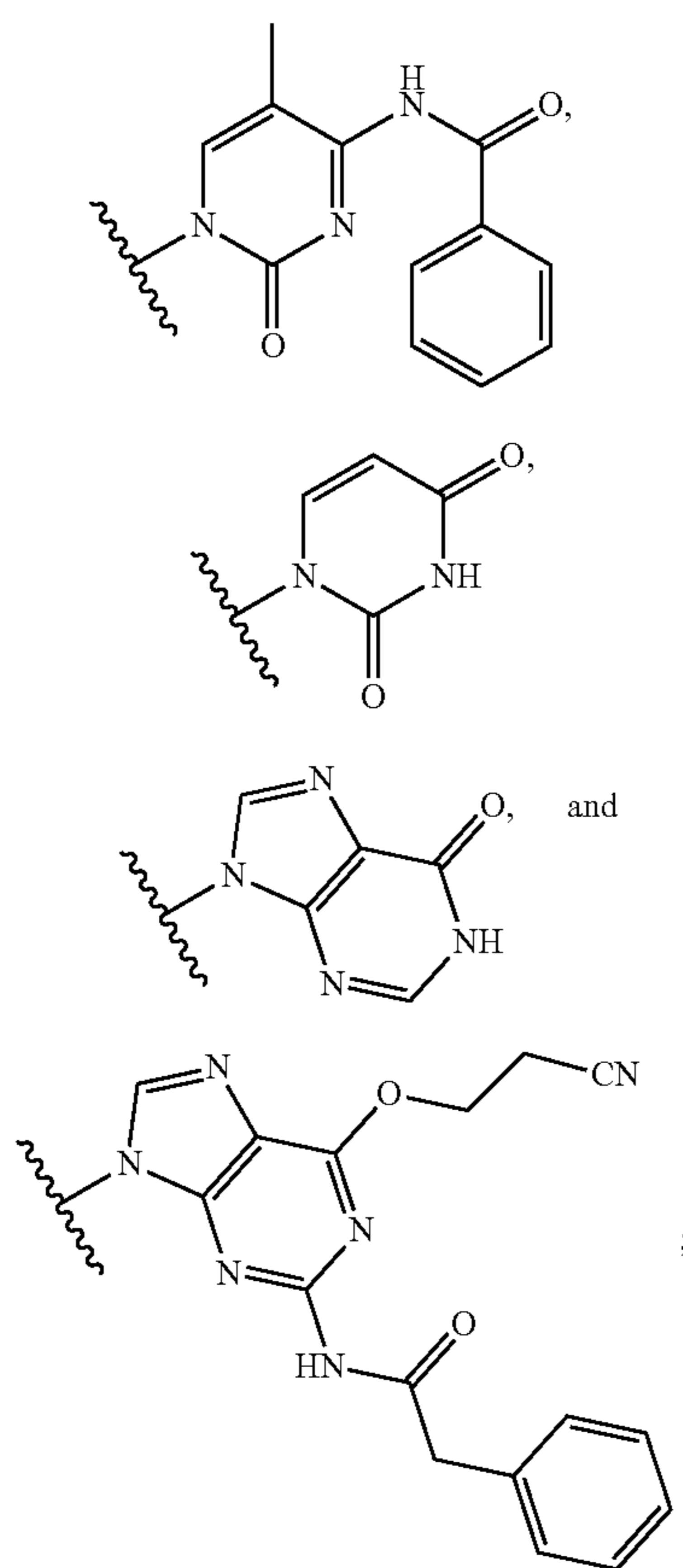
(T)



(PA)

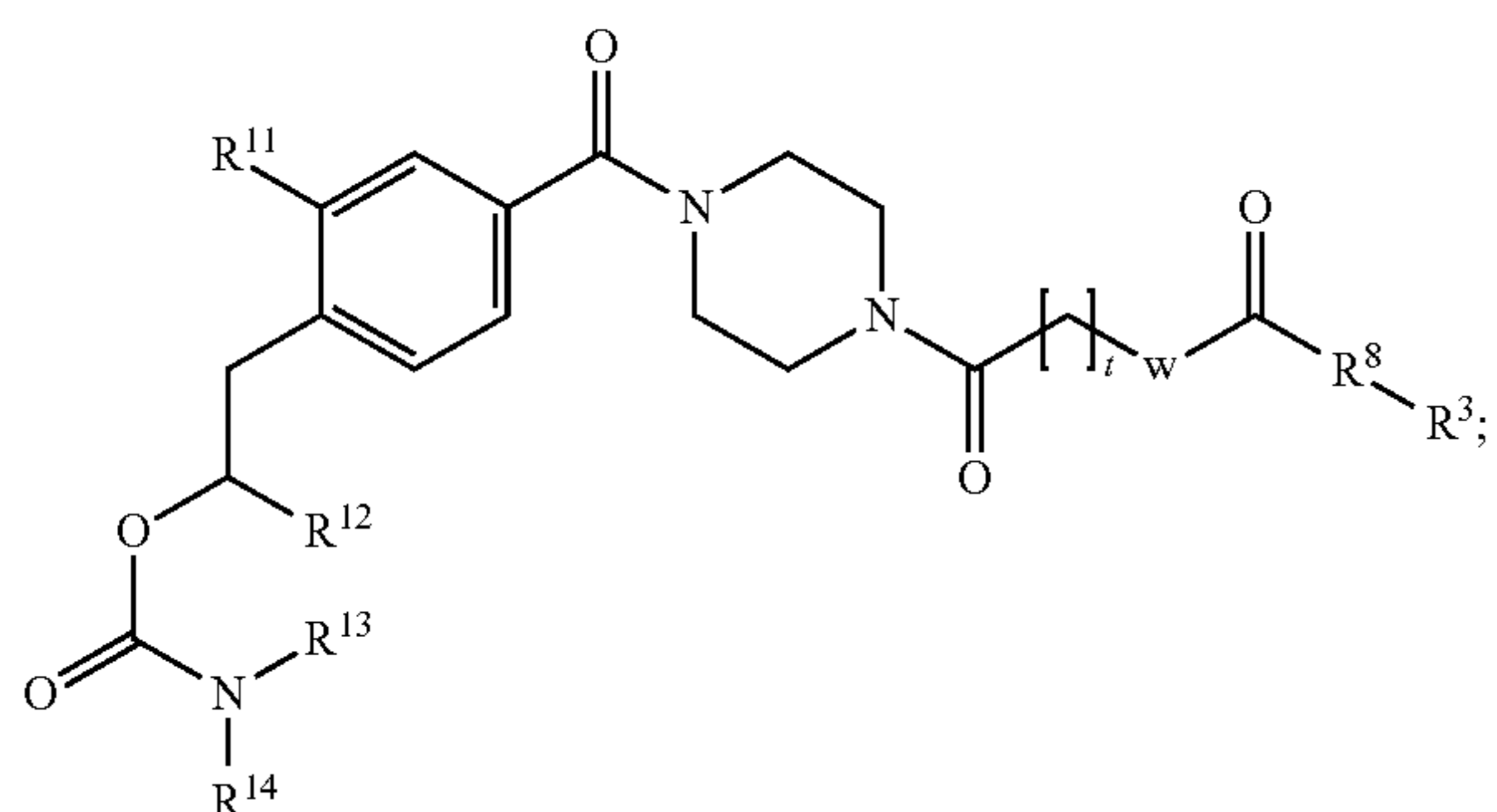
225

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R³ is, independently at each occurrence, selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

R⁹ and R¹⁰ are, independently, C₁₋₆-alkyl, or R⁹ and R¹⁰, together with the atoms to which they are attached, form a cyclic or polycyclic ring comprising 4-10 carbon atoms to form a compound of Formula (B3):



wherein

w is selected from O or C;

t is 1, 2, 3, 4, or 5;

R¹¹ is selected from the group consisting of halo, CN, and NO₂;

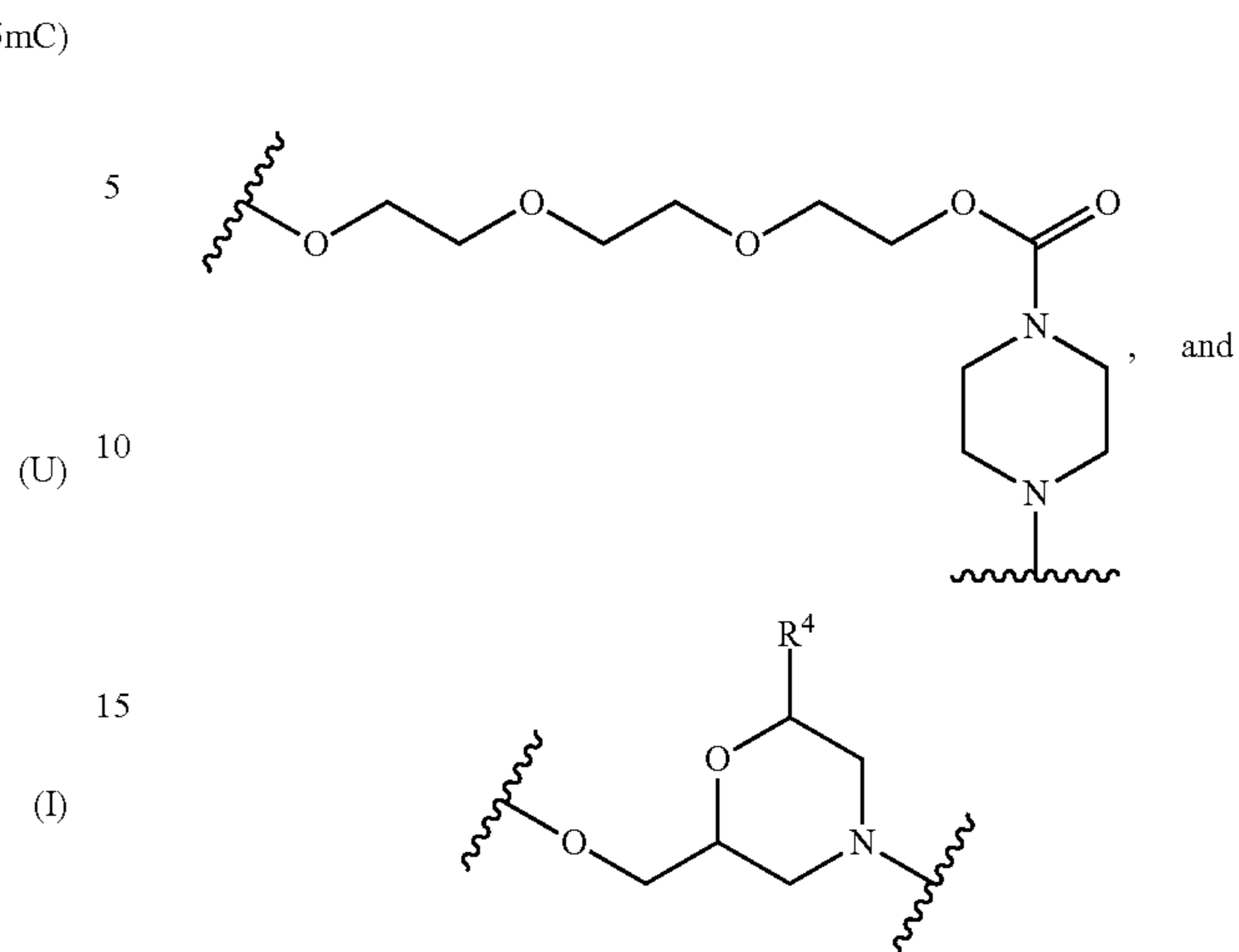
R¹² is C₁₋₆-alkyl;

R¹³ is H or C₁₋₆-alkyl;

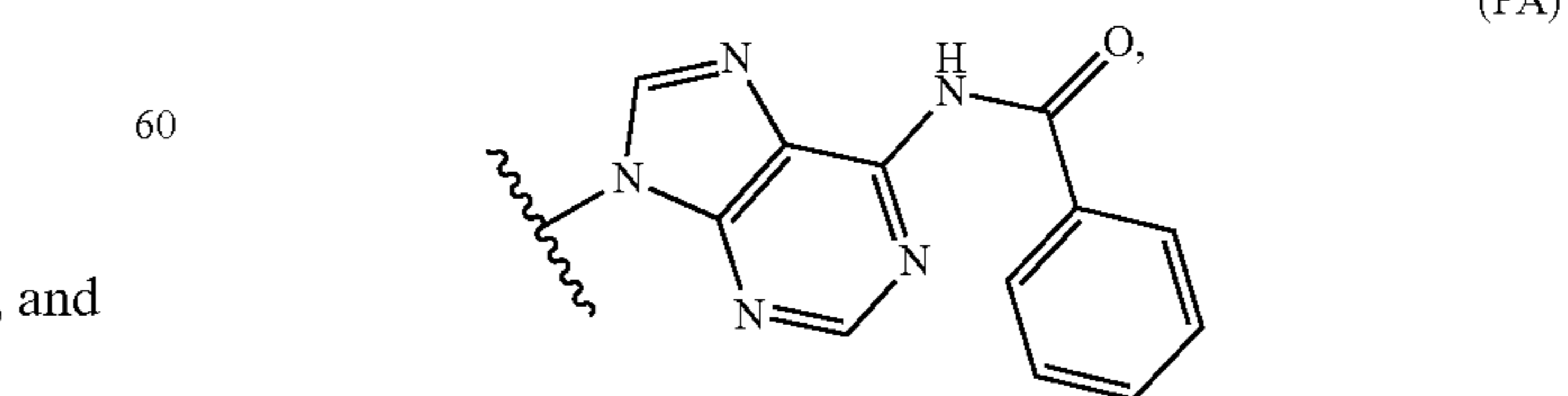
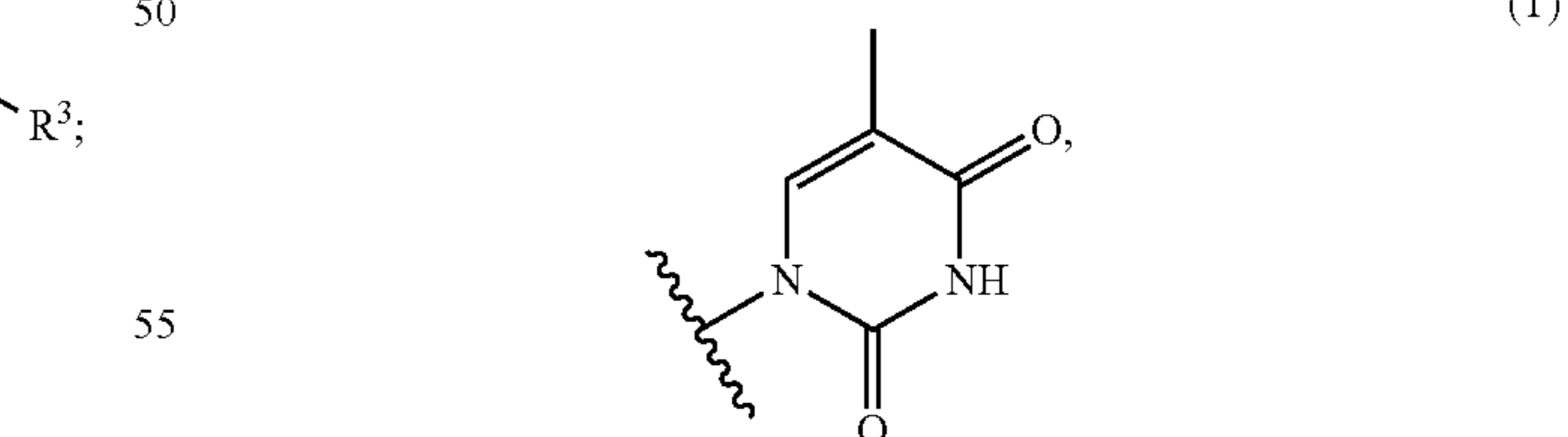
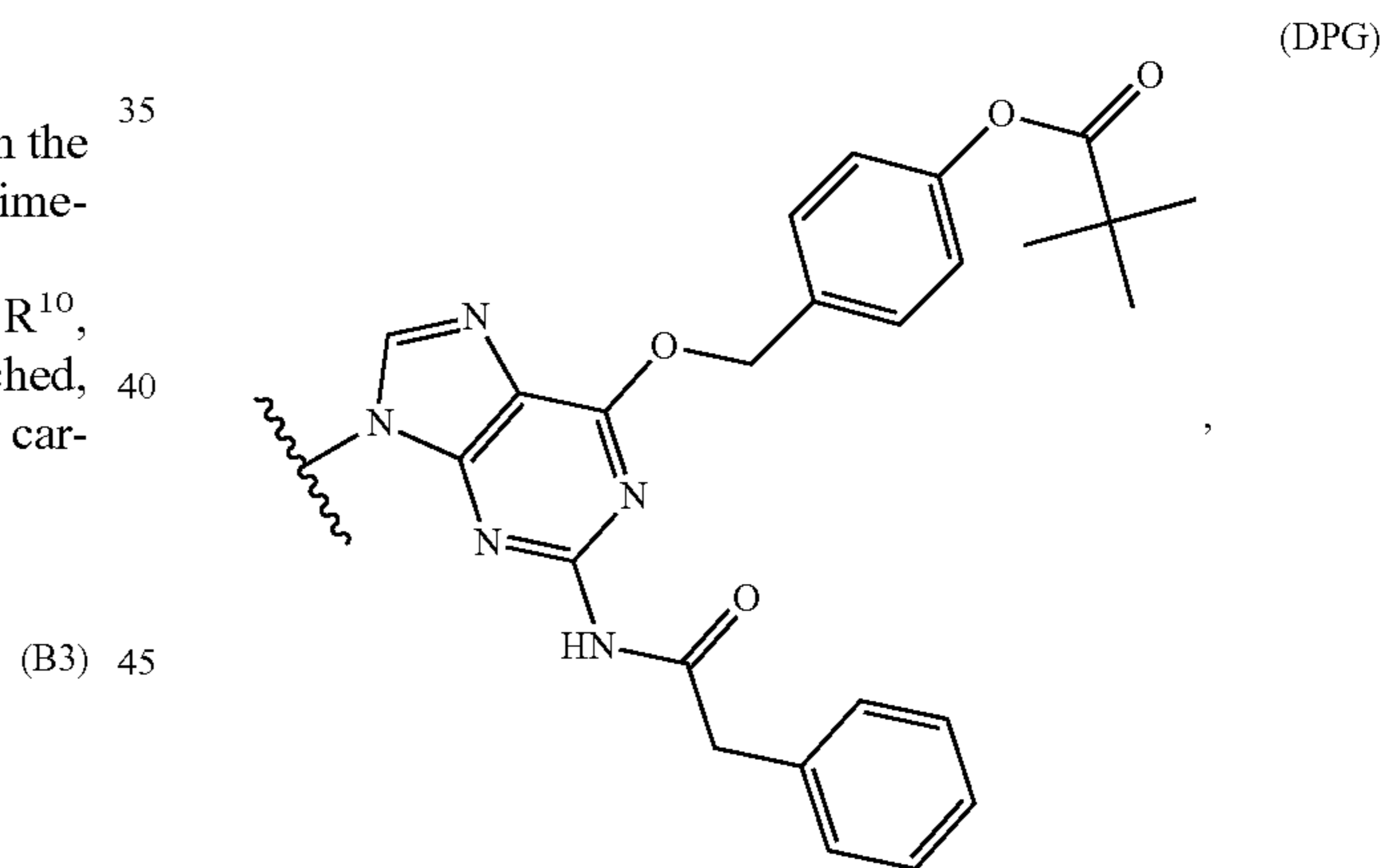
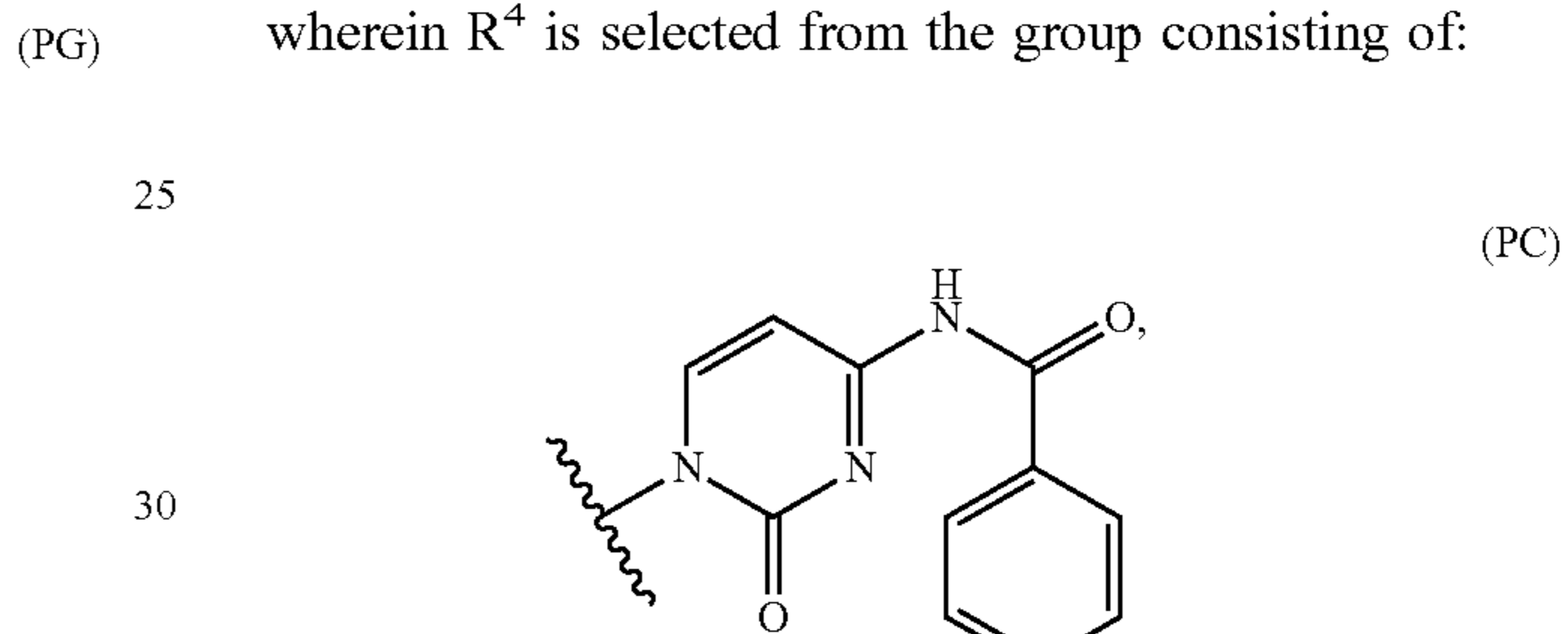
R¹⁴ is a first linker bound to a support-medium;

226

R⁸ is selected from:



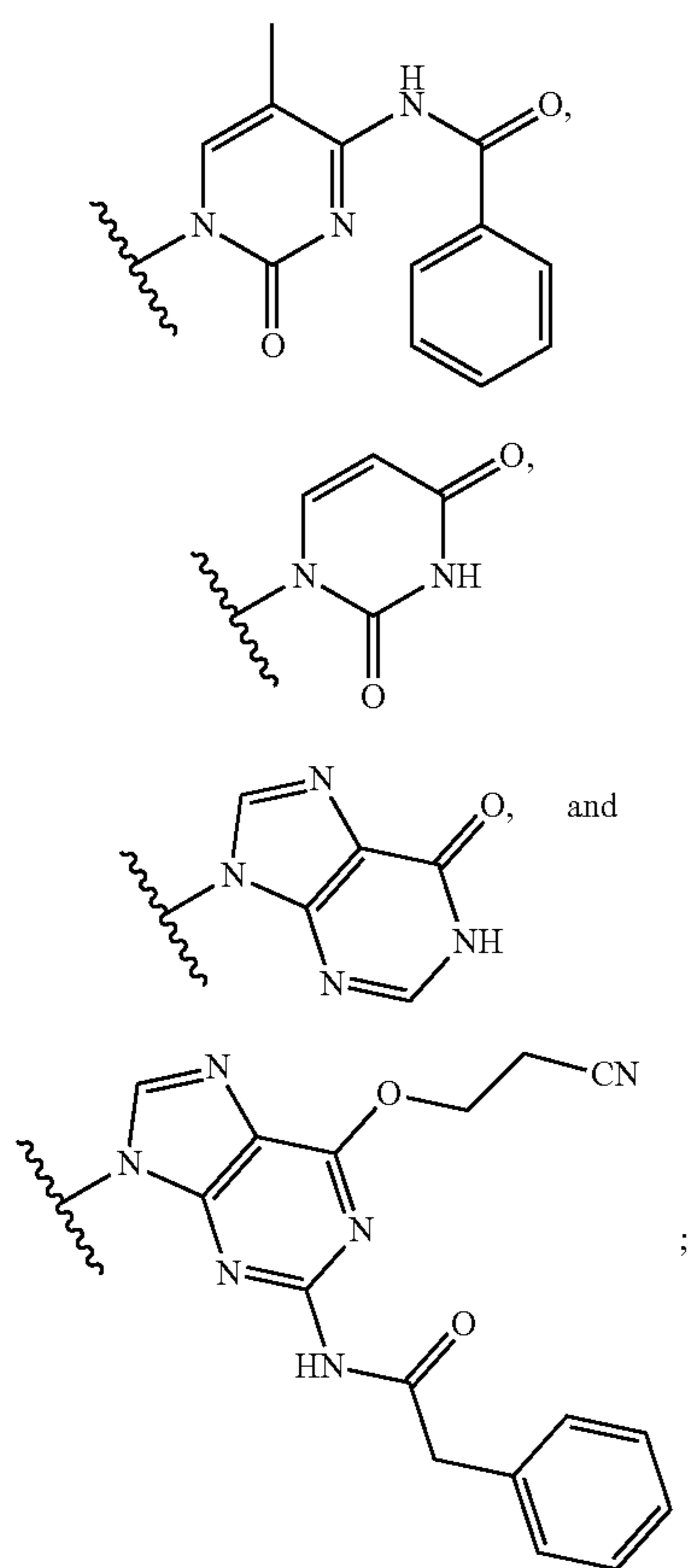
wherein R⁴ is selected from the group consisting of:



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227

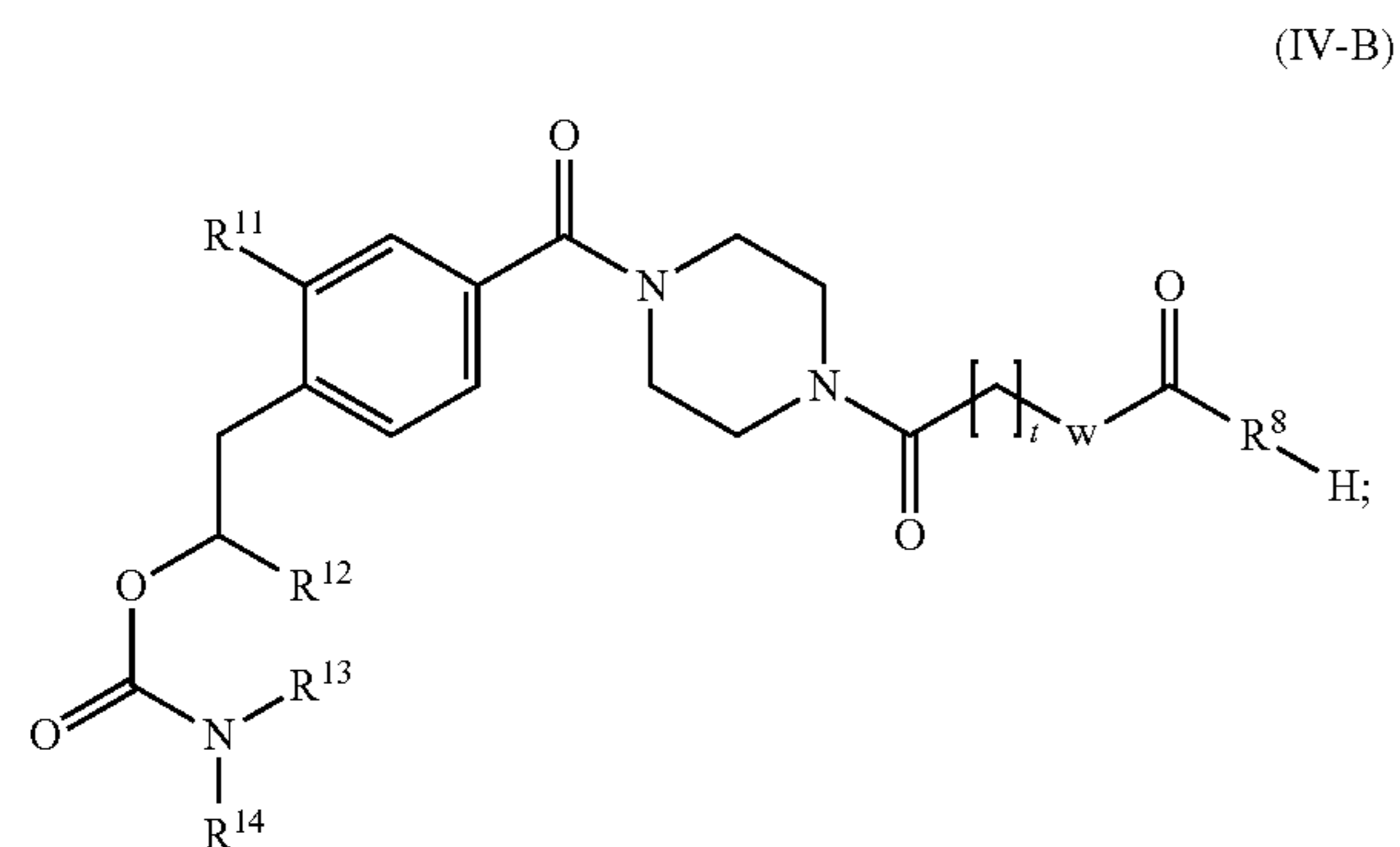
-continued



and

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl;

(c) contacting the compound of Formula (A3) with a



wherein

w is selected from 0 or C;

t is 1, 2, 3, 4, or 5;

R¹¹ is selected from the group consisting of halo, CN, and NO₂;

R¹² is C₁₋₆-alkyl;

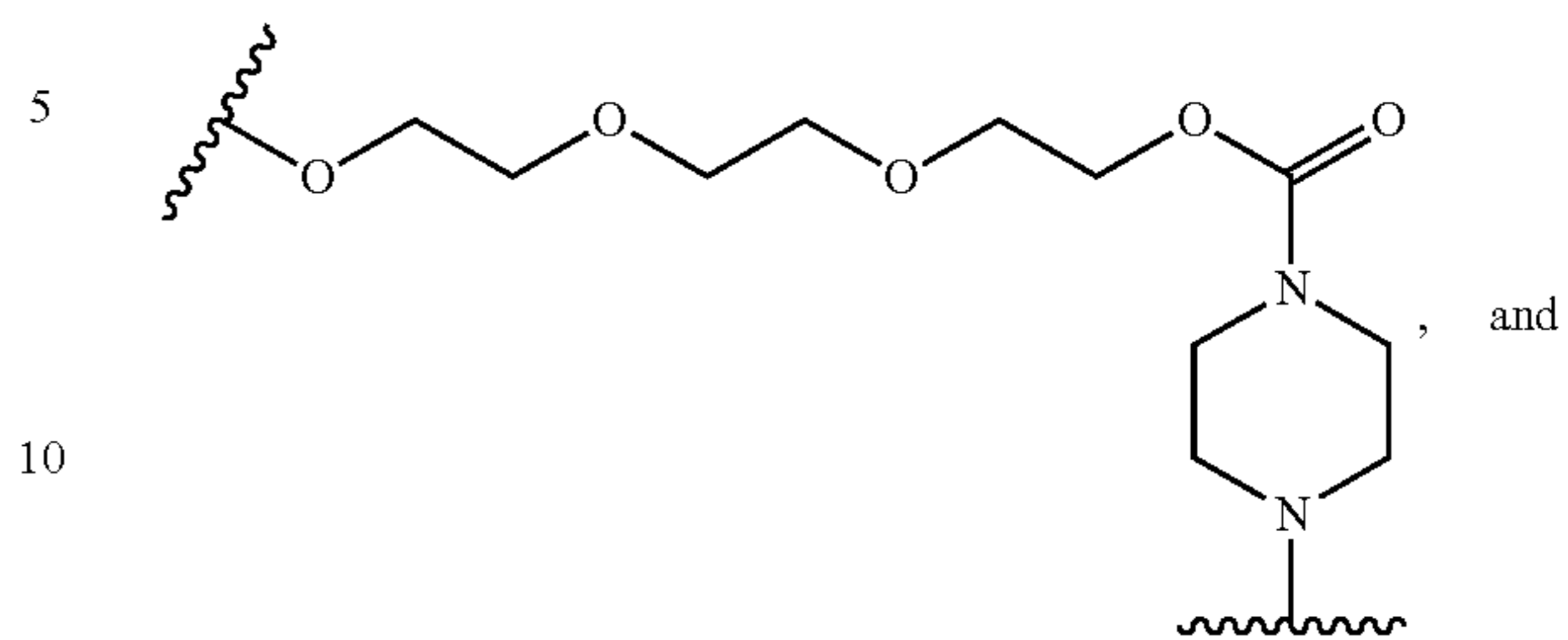
R¹³ is H or C₁₋₆-alkyl;

R¹⁴ is a first linker bound to a support-medium; and

228

R⁸ is selected from:

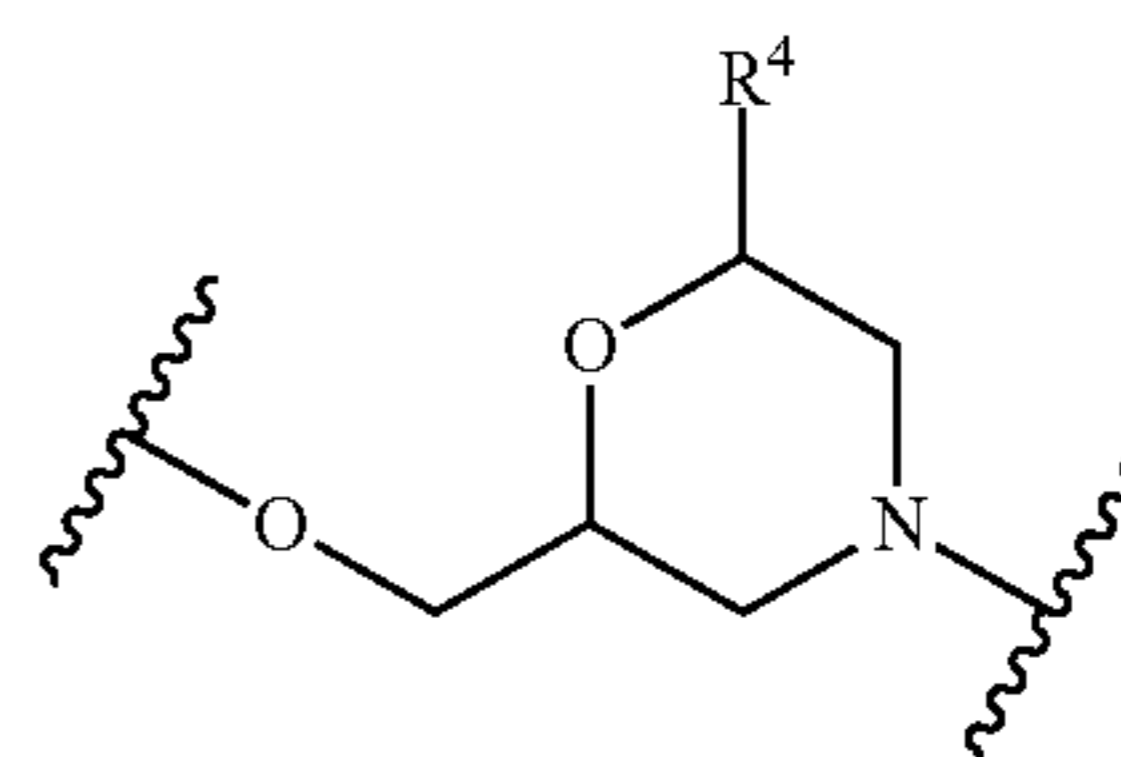
(P5mC)



(U)

15

(I)

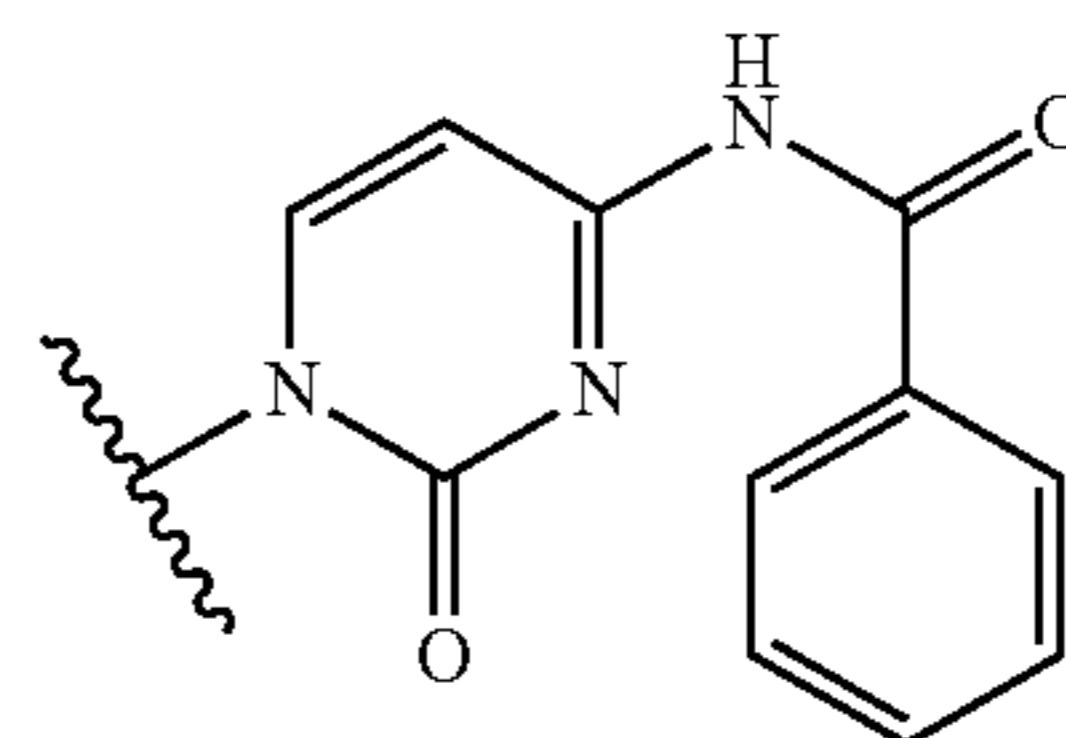


20

(PG)

wherein R⁴ is selected from the group consisting of:

25

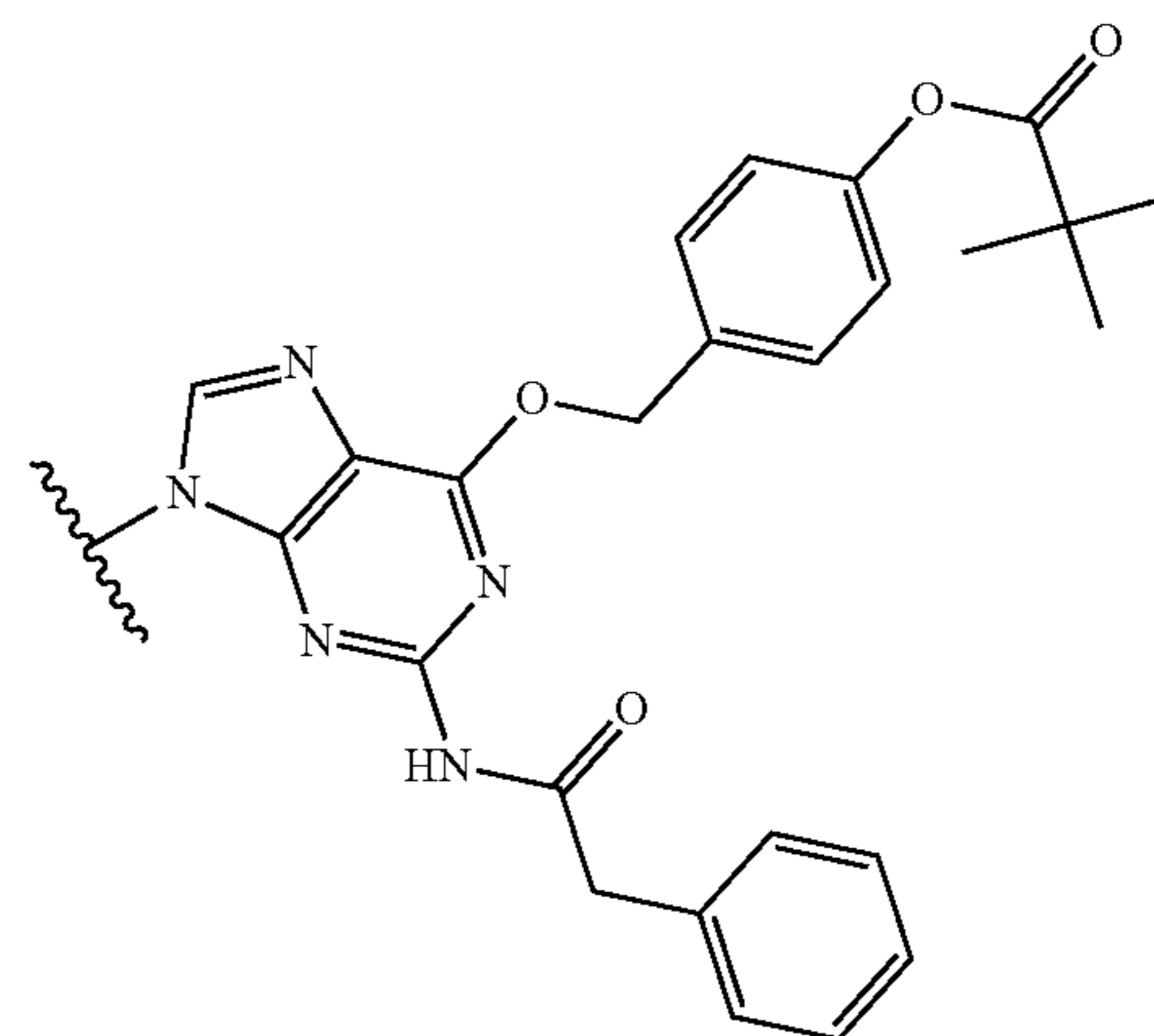


(PC)

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(DPG)

35



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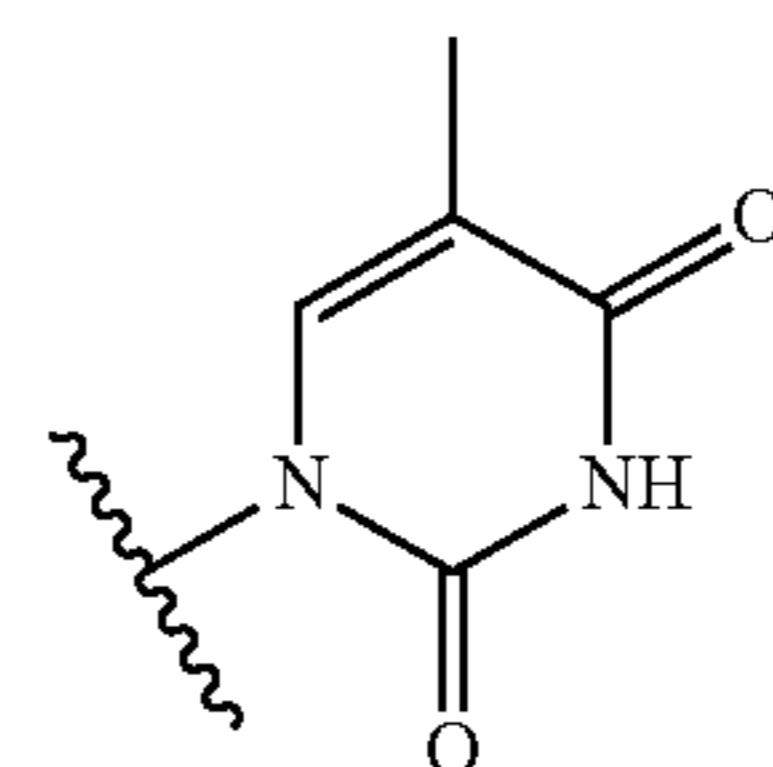
(IV-B)

45

(T)

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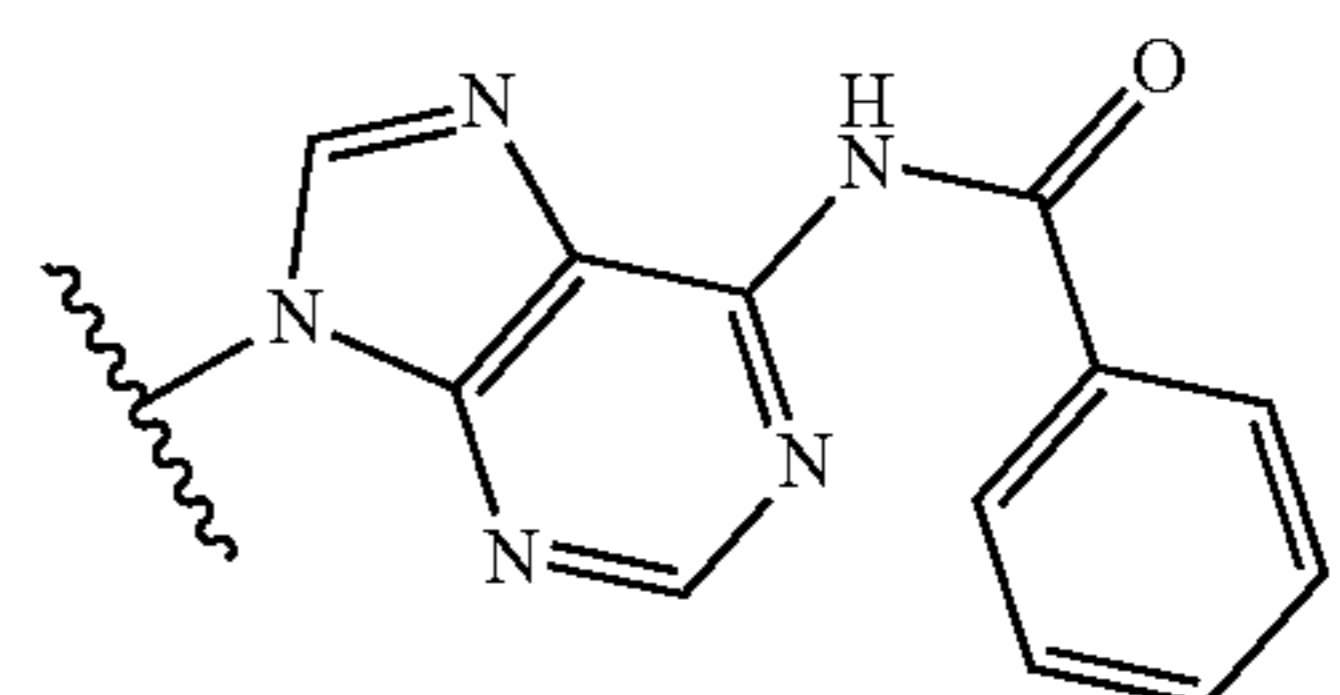
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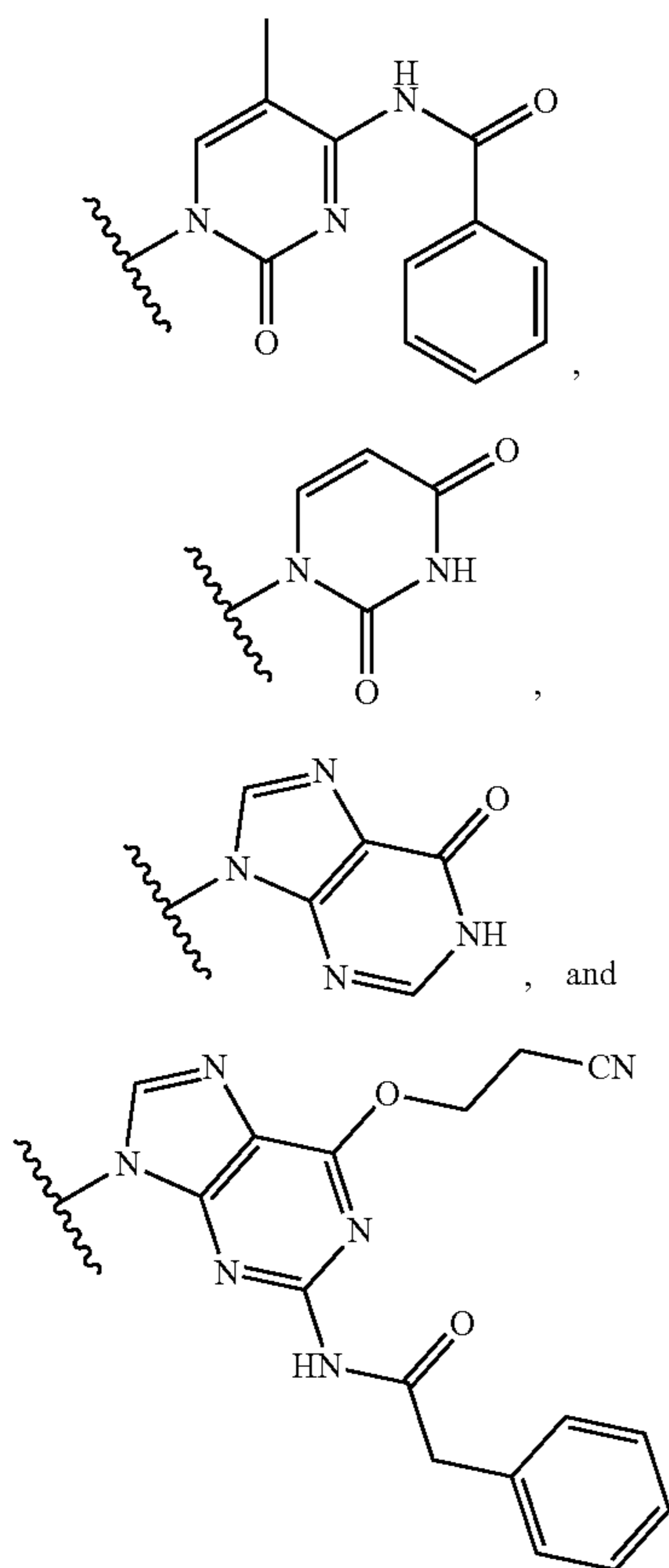
(PA)

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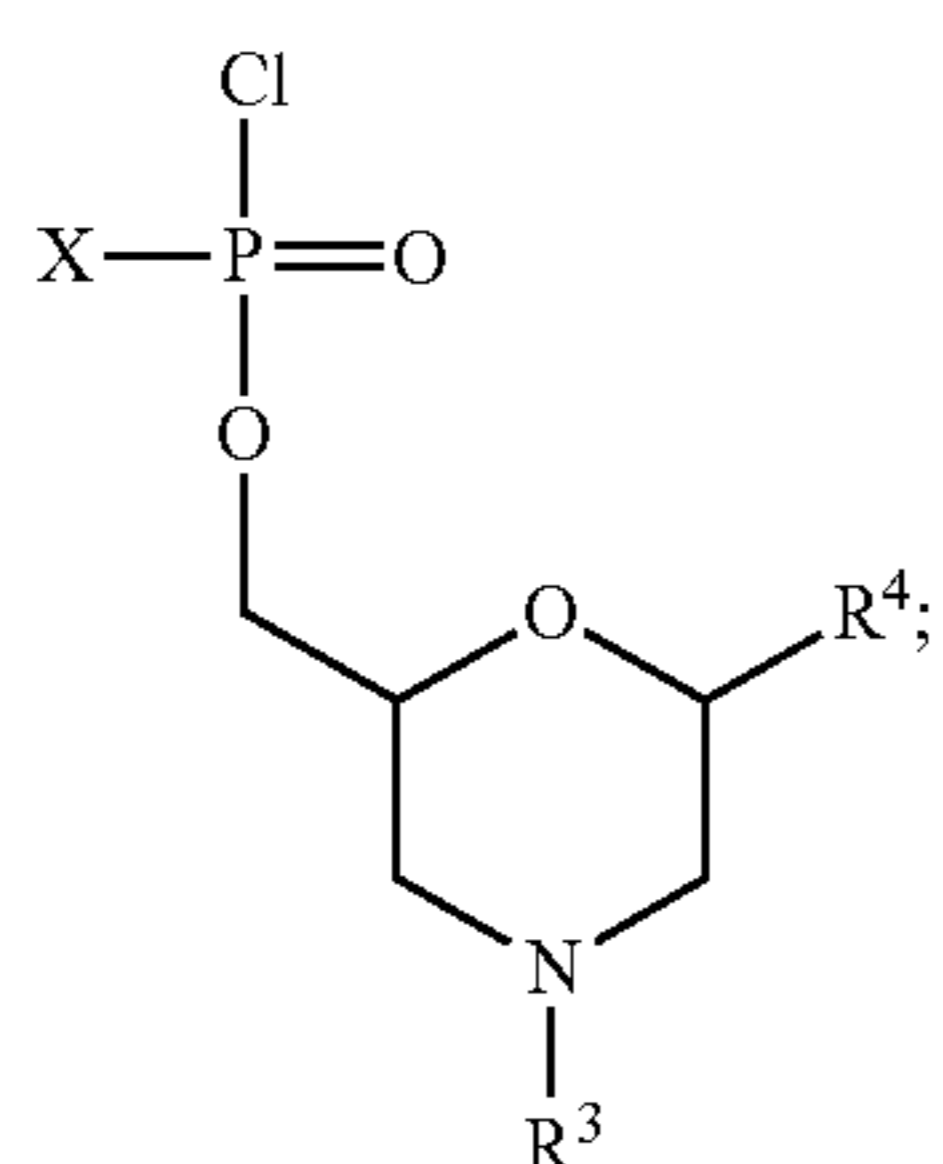


229

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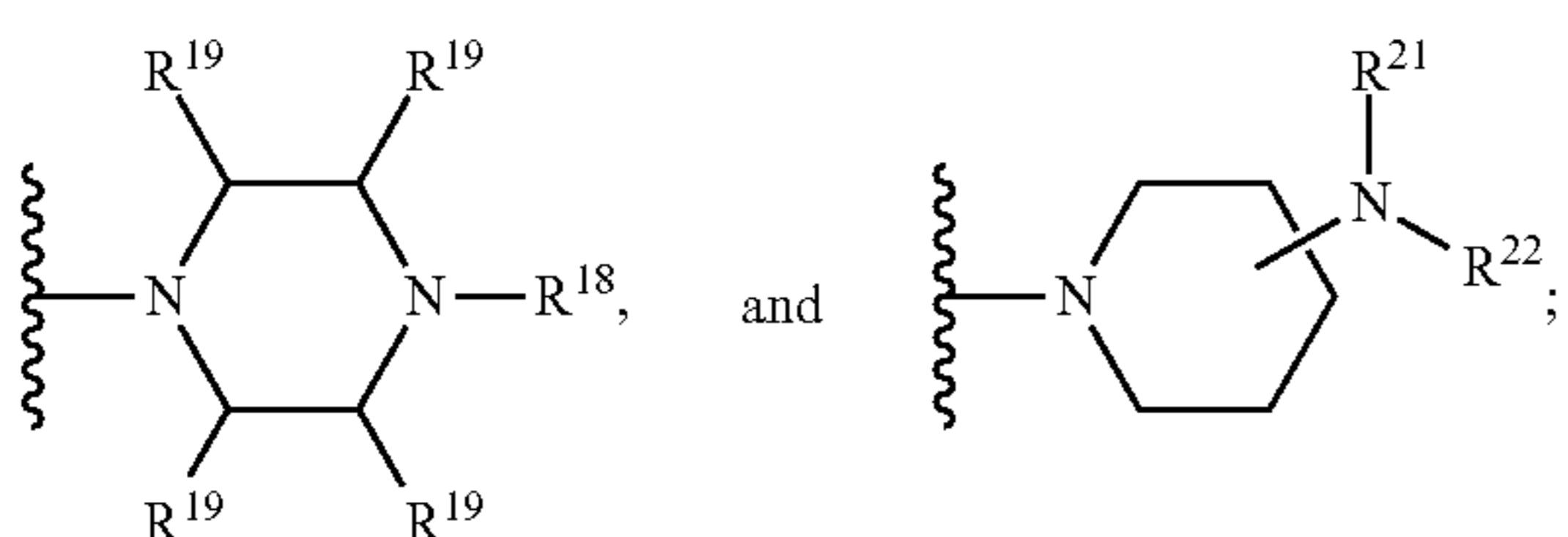


(d) contacting the compound of Formula (IV-B) with a compound of Formula (B4):



wherein

X is, independently at each occurrence, selected from H, OH, $-\text{OCH}_2\text{CH}_2\text{CN}$, $\text{N}(\text{R}^{17})_2$,



R^{17} is, independently for each occurrence, H or C_{1-6} -alkyl;

230

R^{18} is selected from the group consisting of H, C_{1-6} -alkyl, $-\text{C}(=\text{NH})\text{NH}_2$, $-\text{C}(\text{O})(\text{CH}_2)_q\text{NR}^{20}\text{C}(=\text{NH})\text{NH}_2$, and $-\text{C}(\text{O})(\text{CH}_2)_2\text{NHC}(\text{O})(\text{CH}_2)_5\text{NR}^{20}\text{C}(=\text{NH})\text{NH}_2$;

q is 1, 2, 3, 4, or 5;

R^{19} is, independently for each occurrence, H or methyl;

R^{20} is H or C_{1-6} -alkyl;

R^{21} is selected from the group consisting of H, C_{1-6} -alkyl, $\text{C}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})(\text{CH}_2)_r\text{NR}^{23}\text{C}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})\text{CH}(\text{NH}_2)(\text{CH}_2)_3\text{NHC}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})(\text{CH}_2)_2\text{NHC}(\text{O})(\text{CH}_2)_5\text{NR}^{23}\text{C}(=\text{NH})\text{NH}_2$, and $\text{C}(\text{O})\text{CH}(\text{NH}_2)(\text{CH}_2)_4\text{NH}_2$;

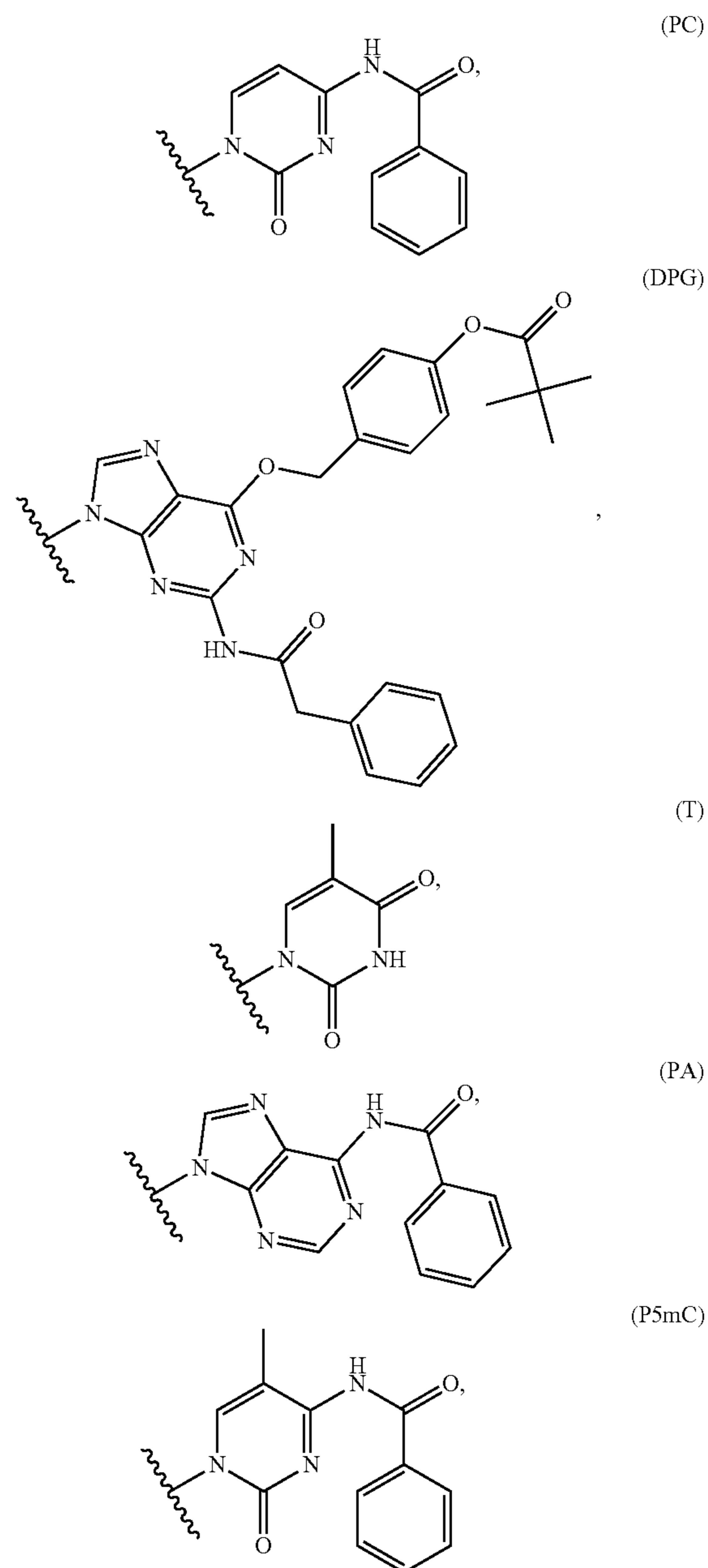
R^{22} is H or C_{1-6} -alkyl;

R^{23} is H or C_{1-6} -alkyl; and

r is 1, 2, 3, 4, or 5;

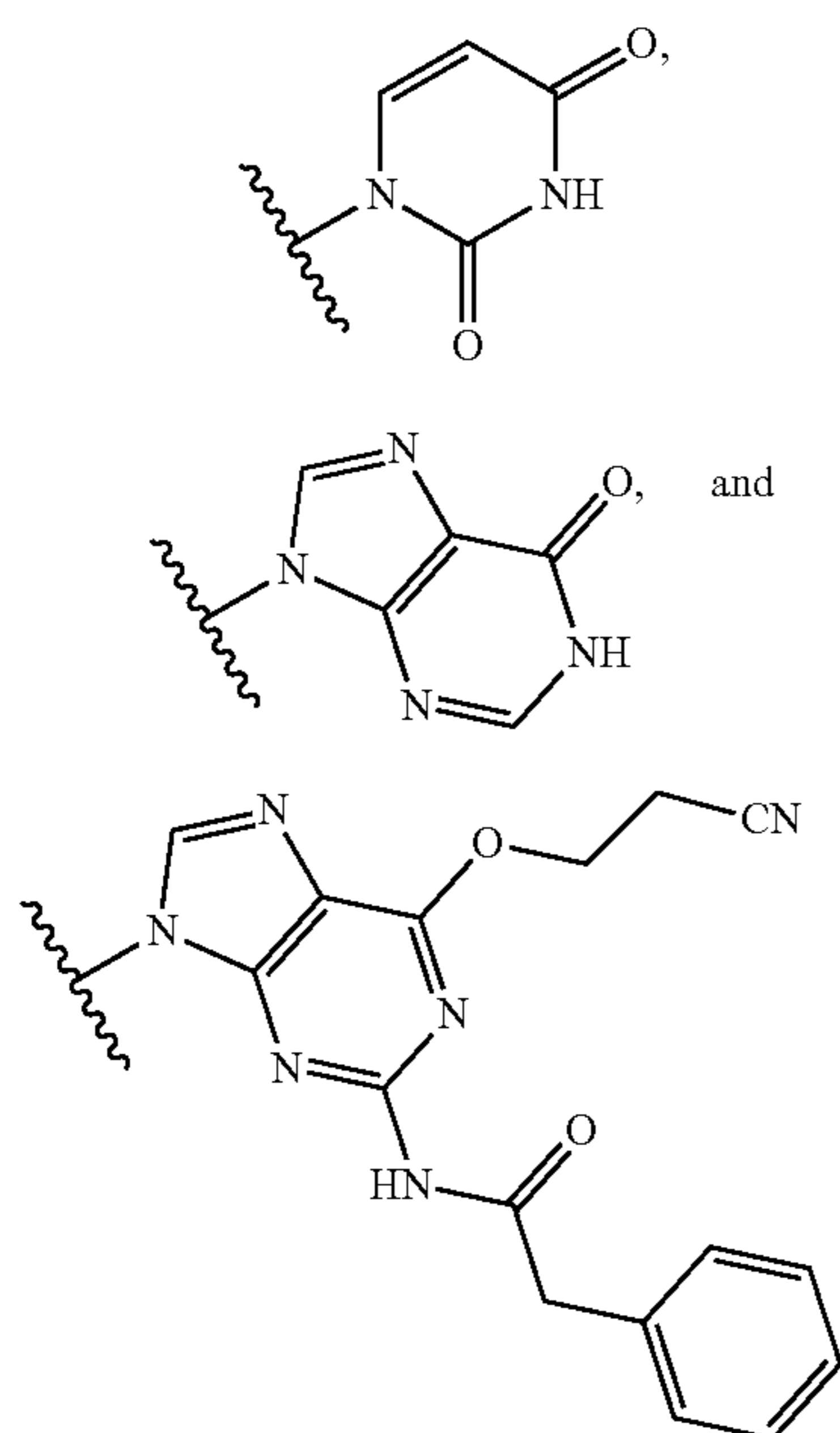
R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

R^4 is selected from the group consisting of:

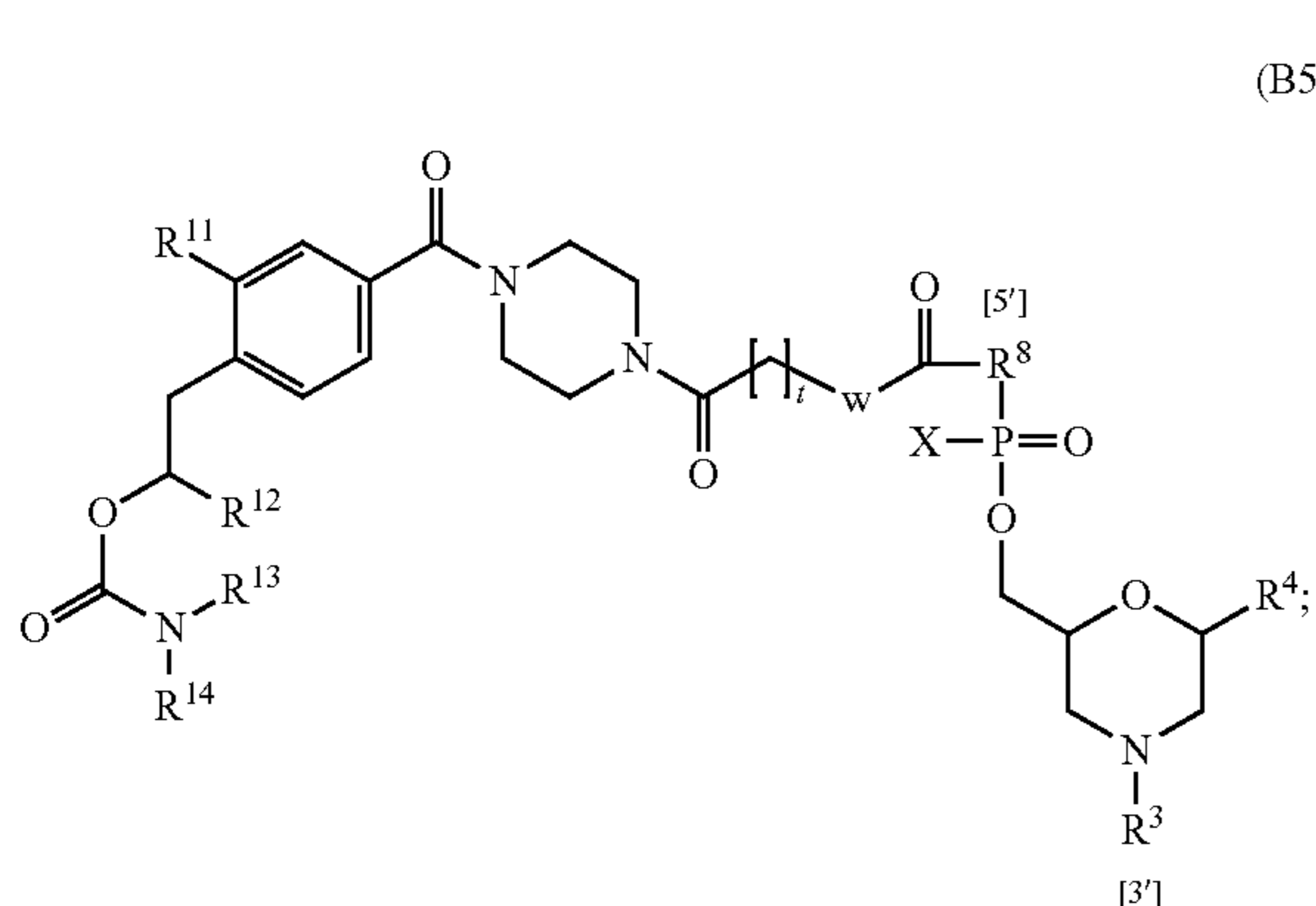


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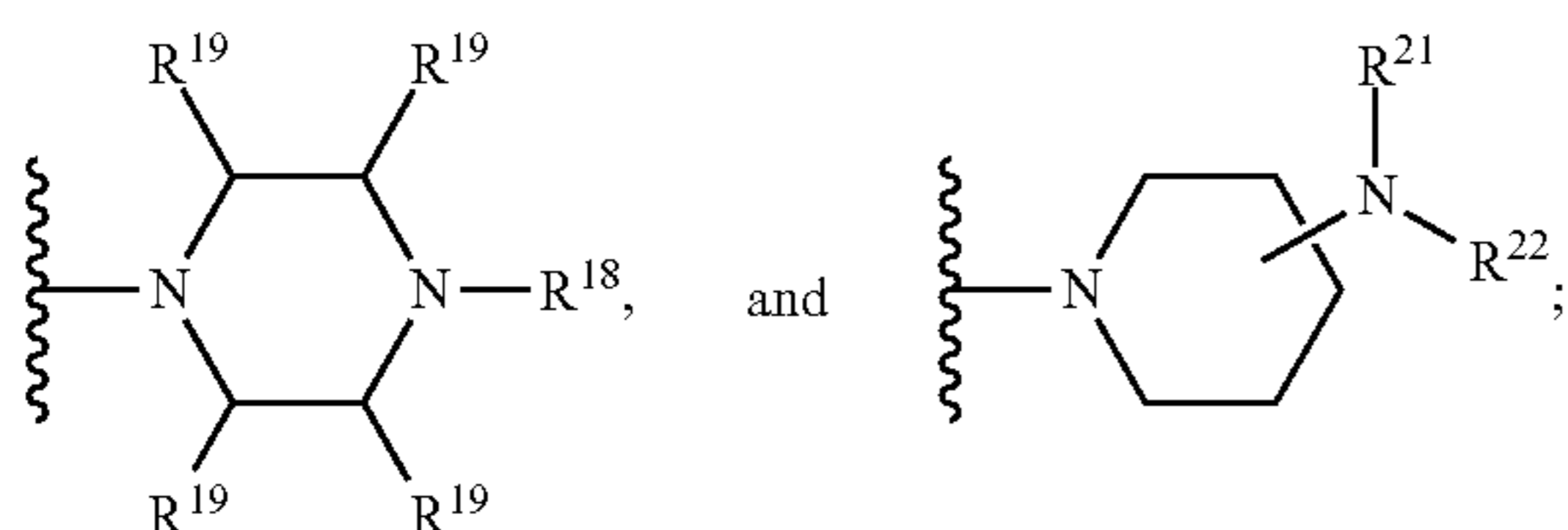
to form a compound of Formula (B5):



wherein:

w is selected from O or C;

t is 1, 2, 3, 4, or 5;

R¹¹ is selected from the group consisting of halo, CN, and NO₂;R¹² is C₁₋₆-alkyl;R¹³ is H or C₁₋₆-alkyl;R¹⁴ is a first linker bound to a support-medium;X is, independently at each occurrence, selected from H, OH, —OCH₂CH₂CN, N(R¹⁷)₂,

232

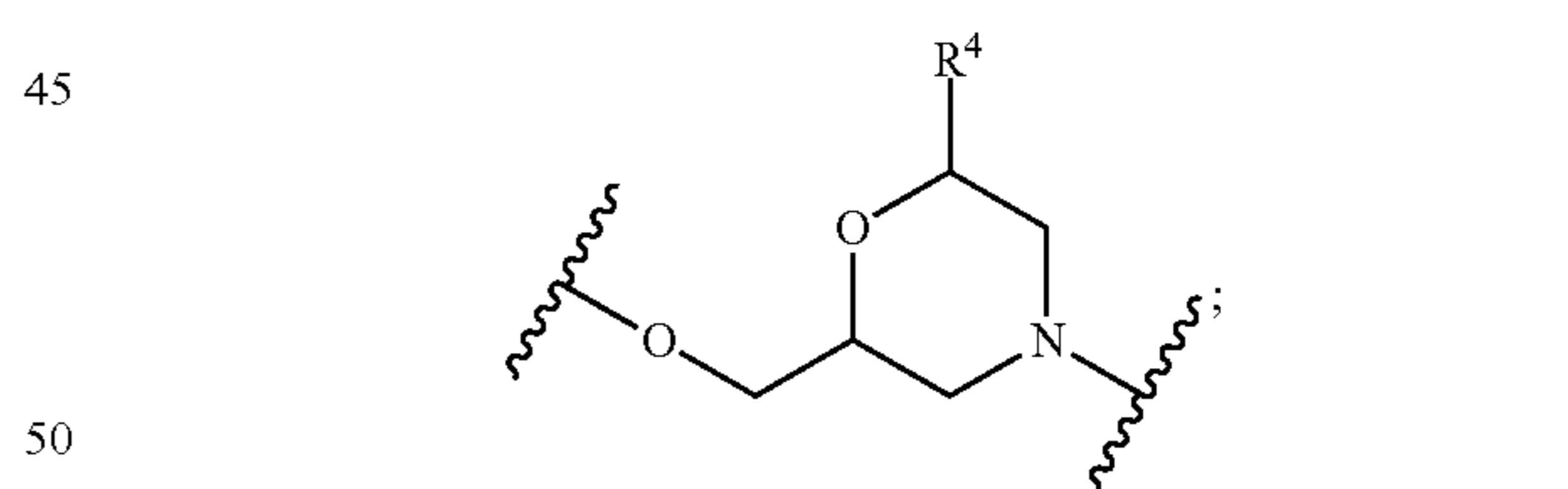
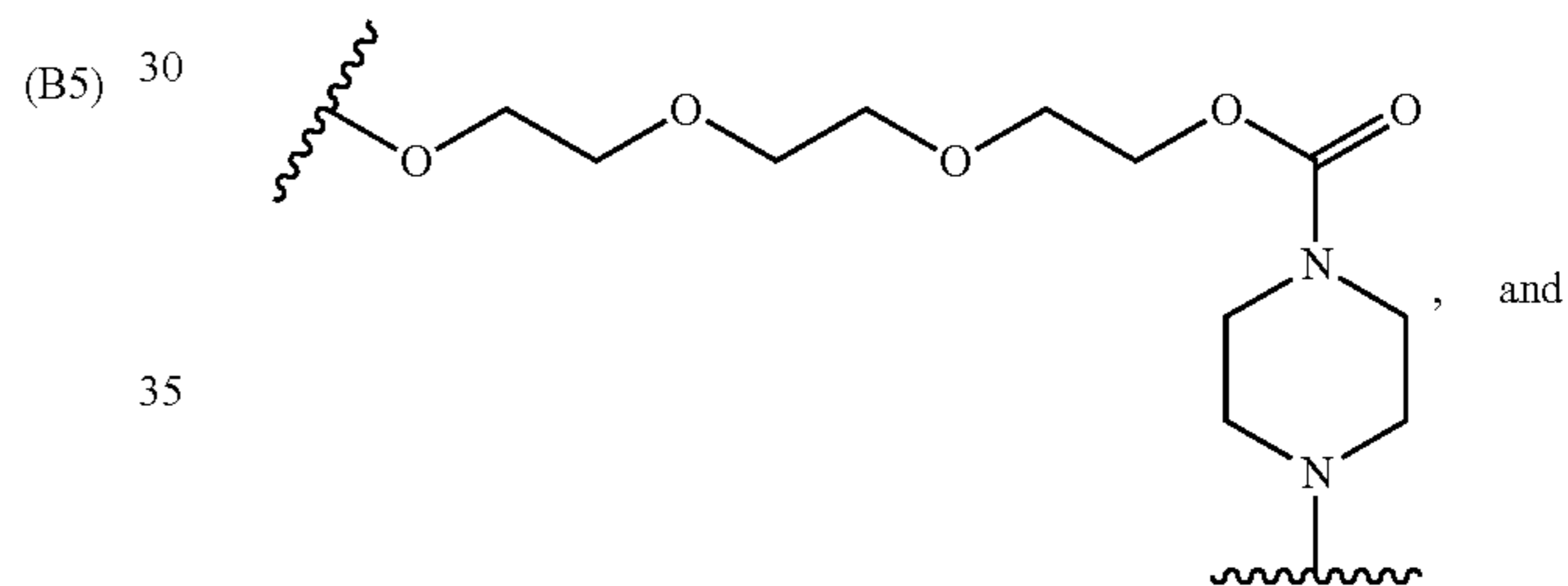
wherein:

(U) R¹⁷ is, independently for each occurrence, H or C₁₋₆-alkyl;5 R¹⁸ is selected from the group consisting of H, C₁₋₆-alkyl, —C(=NH)NH₂, —C(O)(CH₂)_qNR²⁰C(=NH)NH₂, and —C(O)(CH₂)₂NHC(O)(CH₂)₅NR²⁰C(=NH)NH₂;

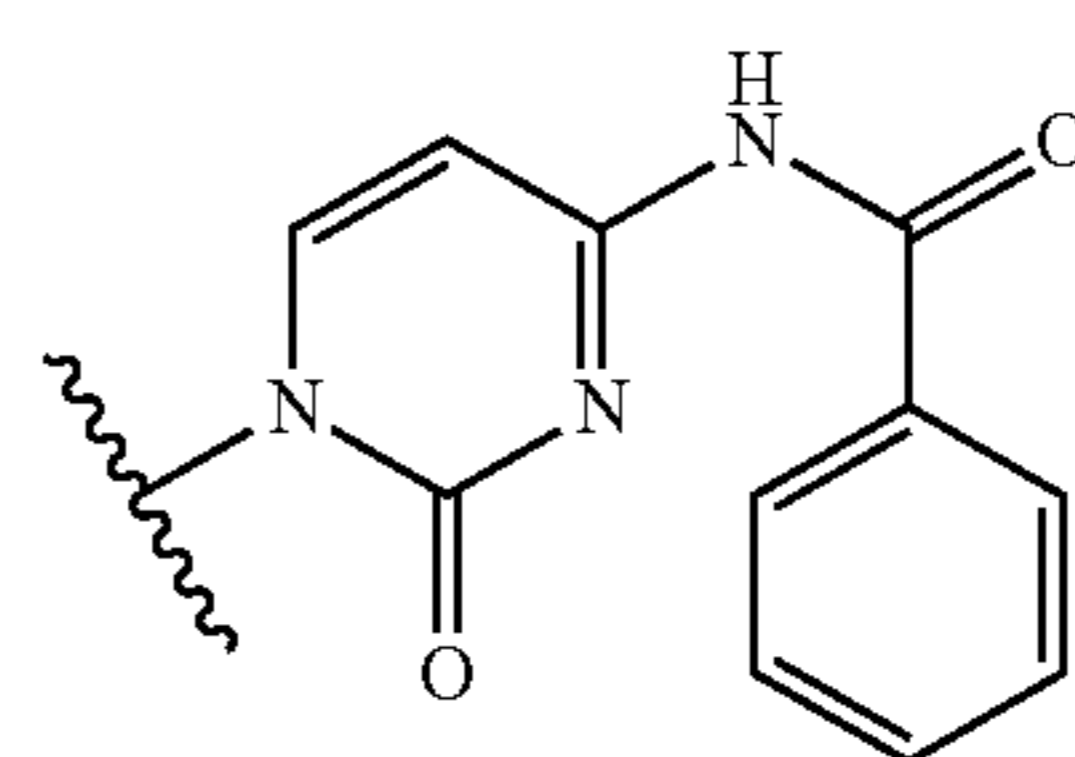
(I) q is 1, 2, 3, 4, or 5;

10 R¹⁹ is, independently for each occurrence, H or methyl;R²⁰ is H or C₁₋₆-alkyl;(PG) 15 R²¹ is selected from the group consisting of H, C₁₋₆-alkyl, C(=NH)NH₂, C(O)(CH₂)_rNR²³C(=NH)NH₂, C(O)CH(NH₂)(CH₂)₃NHC(=NH)NH₂, C(O)(CH₂)₂NHC(O)(CH₂)₅NR²³C(=NH)NH₂, and C(O)CH(NH₂)(CH₂)₄NH₂;R²² is H or C₁₋₆-alkyl;20 R²³ is H or C₁₋₆-alkyl; and

r is 1, 2, 3, 4, or 5;

25 R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl,R⁸ is selected from:

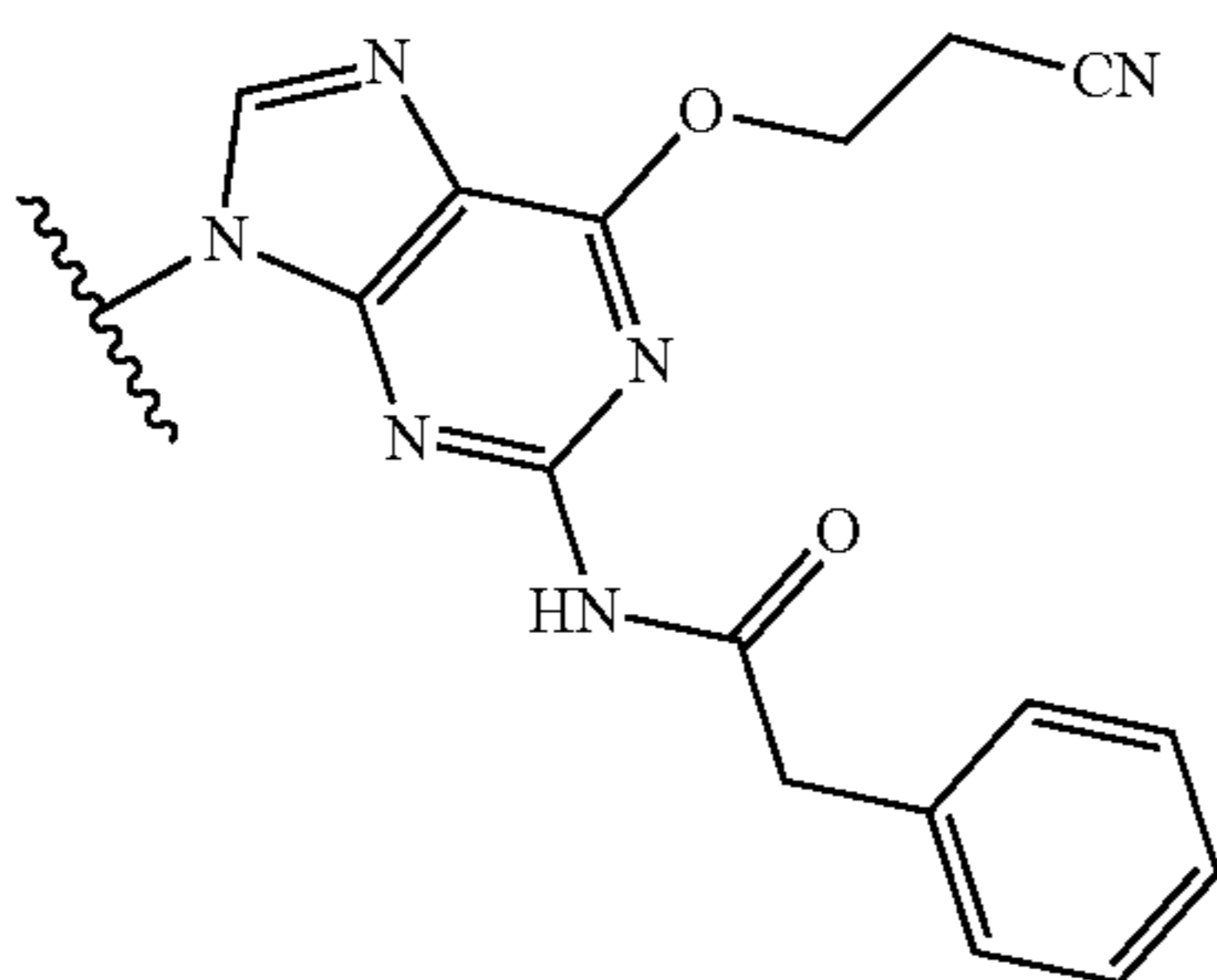
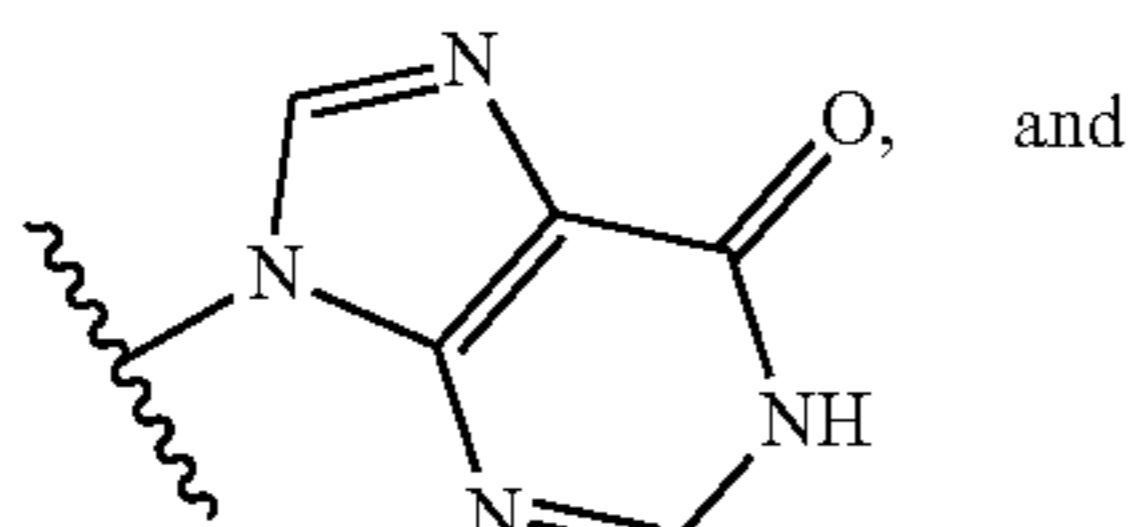
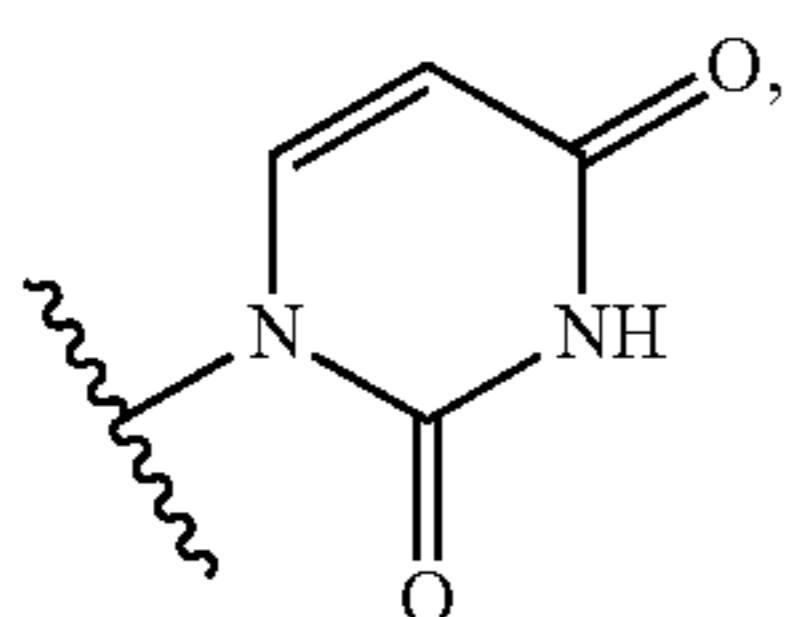
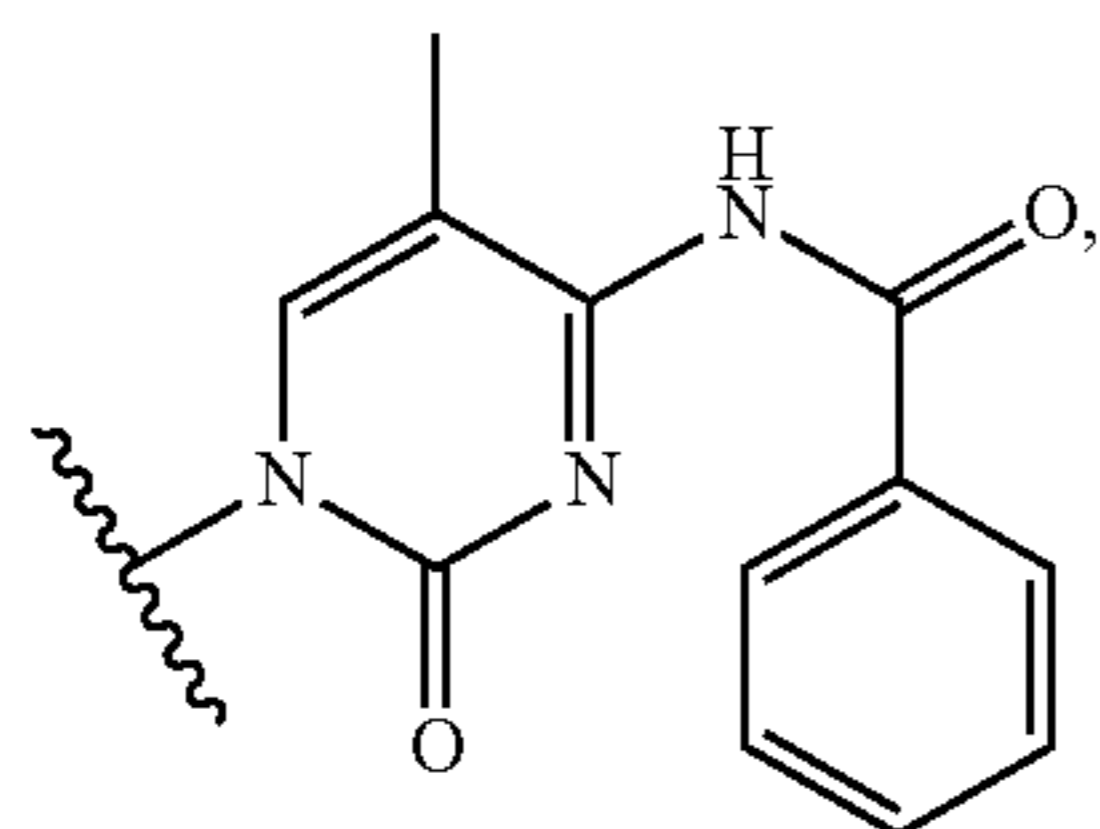
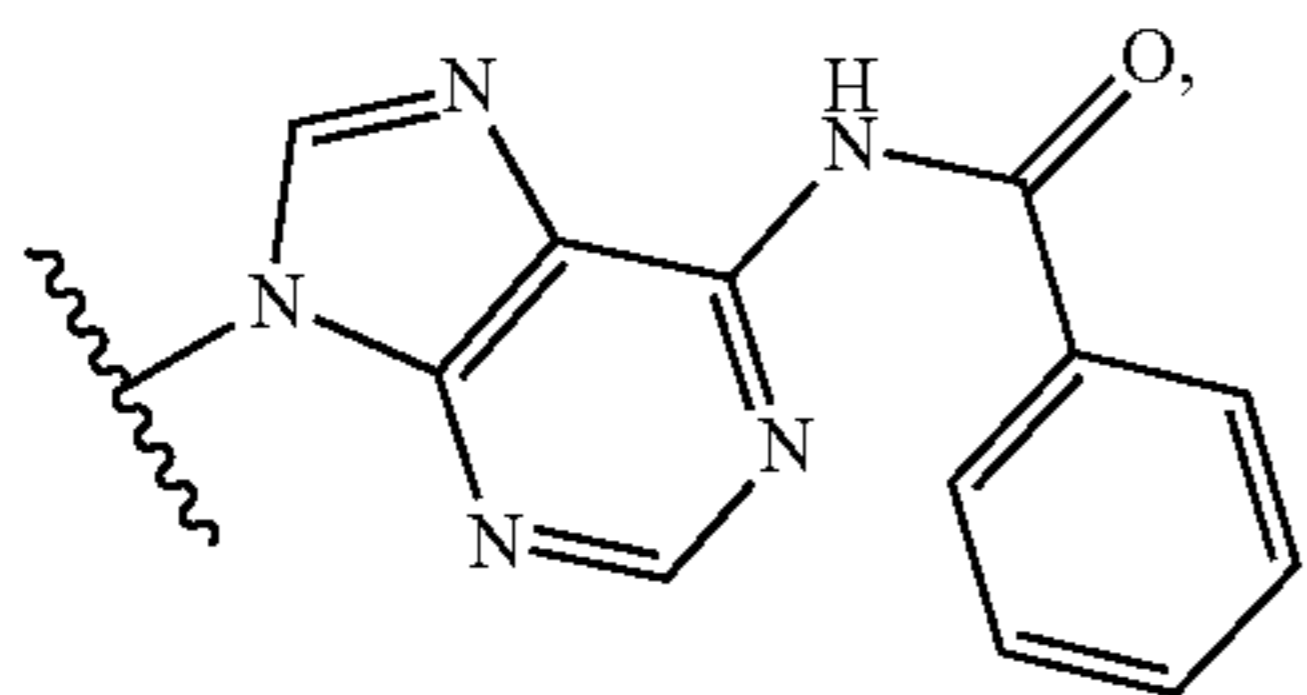
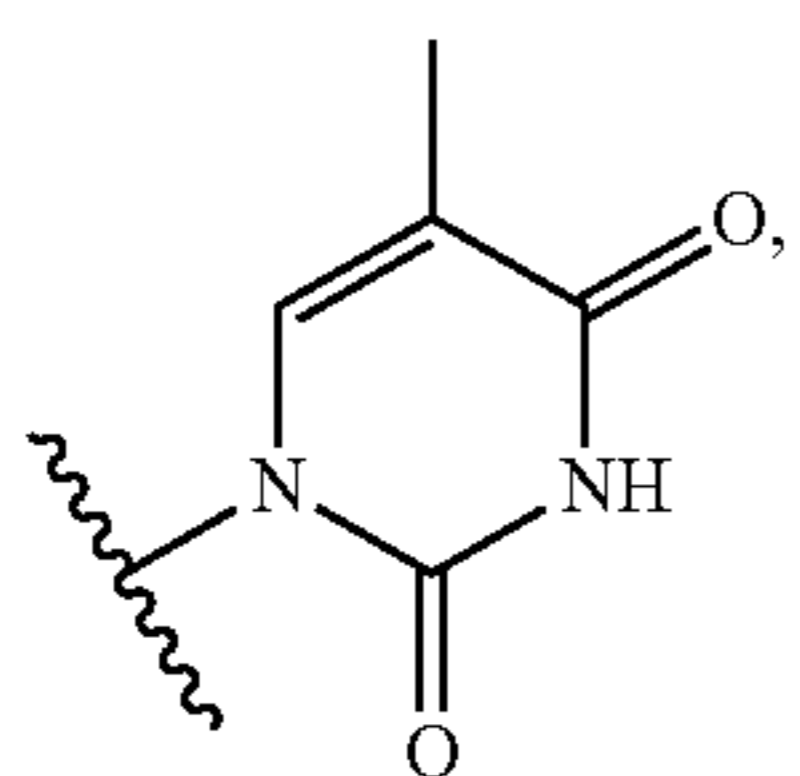
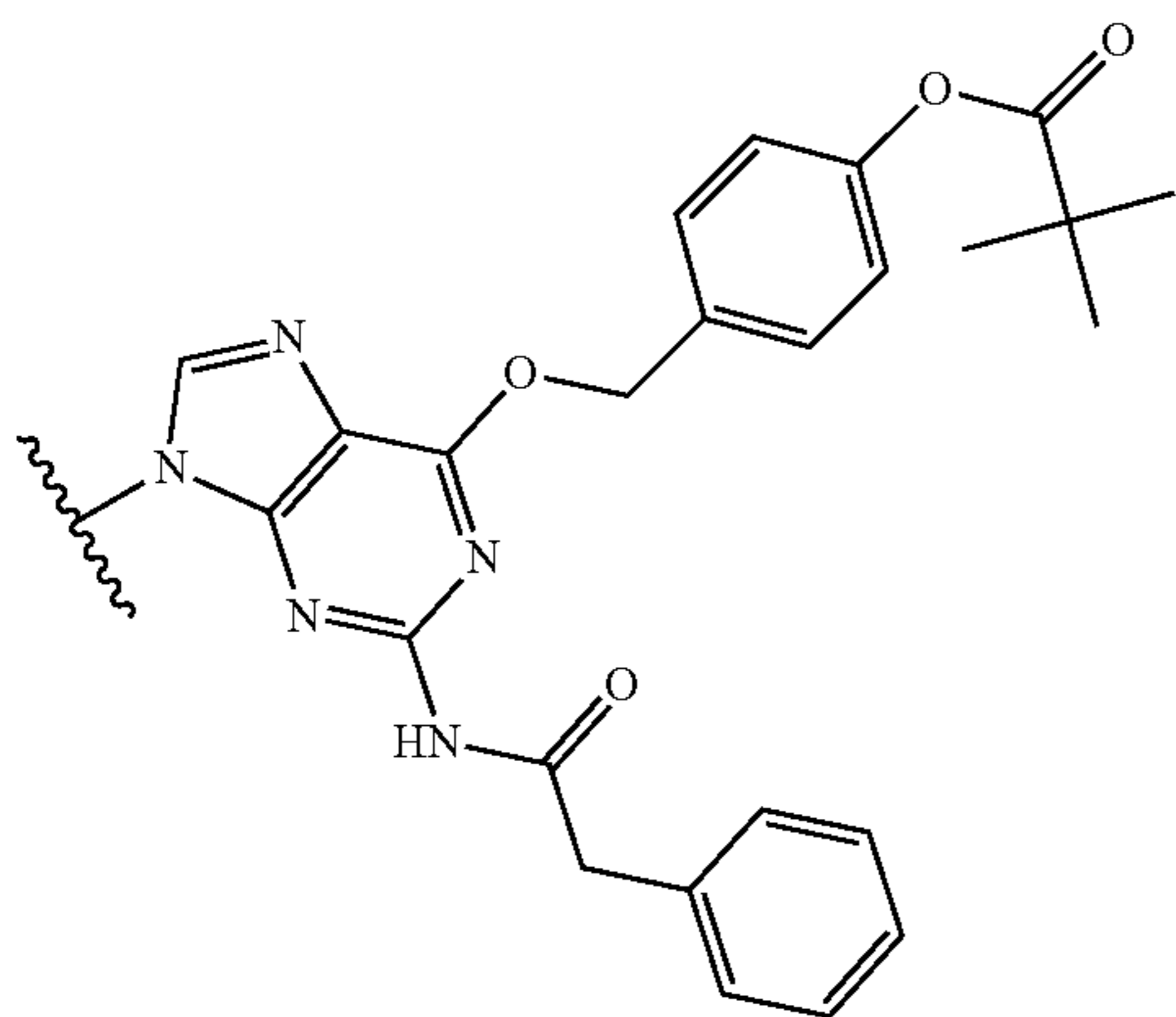
and

R⁴ is, independently at each occurrence, selected from:

(PC)

233

-continued



234

(e) performing n-1 iterations of the sequential steps of:

(e1) contacting the product formed by the immediately prior step with a deblocking agent; and

(e2) contacting the compound formed by the immediately prior step with a compound of Formula (B8):

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(T)

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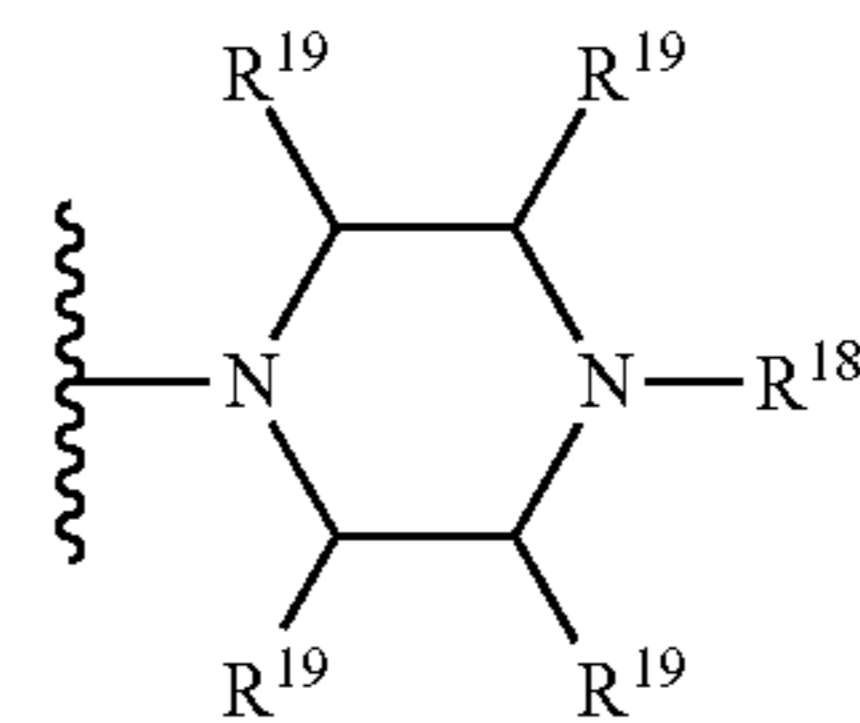
wherein:

X is selected from H, OH, $-\text{OCH}_2\text{CH}_2\text{CN}$, $\text{N}(\text{R}^{17})_2$,

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(PA)

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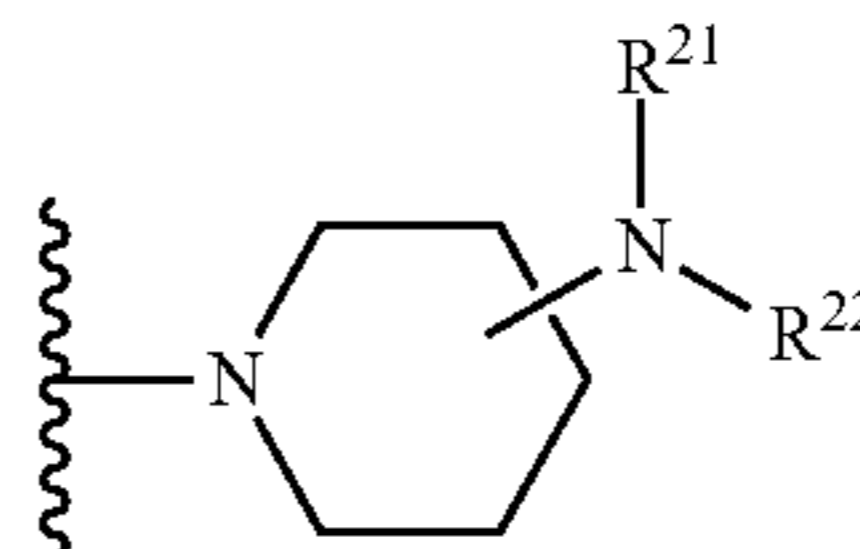
(P5mC)

and

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(U)



wherein

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R^{17} is, independently for each occurrence, H or C_{1-6} -alkyl;

R^{18} is selected from the group consisting of H, C_{1-6} -alkyl, $-\text{C}(=\text{NH})\text{NH}_2$,

(I)

$-\text{C}(\text{O})(\text{CH}_2)_q\text{NR}^{20}\text{C}(=\text{NH})\text{NH}_2$,

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and $-\text{C}(\text{O})(\text{CH}_2)_2\text{NHC}(\text{O})(\text{CH}_2)_5\text{NR}^{20}\text{C}(=\text{NH})\text{NH}_2$;

q is 1, 2, 3, 4, or 5;

R^{19} is, independently for each occurrence, H or methyl;

(PG)

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R^{20} is H or C_{1-6} -alkyl;

R^{21} is selected from the group consisting of H, C_{1-6} -alkyl, $\text{C}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})(\text{CH}_2)_r\text{NR}^{23}\text{C}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})\text{CH}(\text{NH}_2)(\text{CH}_2)_3\text{NHC}(=\text{NH})\text{NH}_2$, $\text{C}(\text{O})(\text{CH}_2)_2\text{NHC}(\text{O})(\text{CH}_2)_5\text{NR}^{23}\text{C}(=\text{NH})\text{NH}_2$, and $\text{C}(\text{O})\text{CH}(\text{NH}_2)(\text{CH}_2)_4\text{NH}_2$;

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R^{22} is H or C_{1-6} -alkyl;

R^{23} is H or C_{1-6} -alkyl;

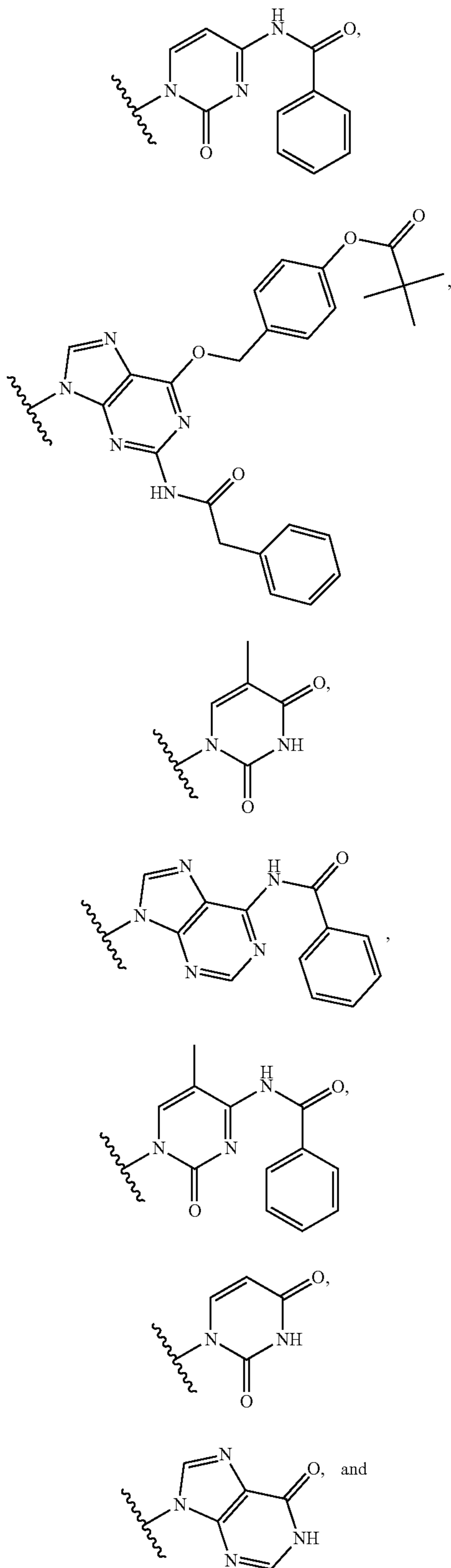
r is 1, 2, 3, 4, or 5;

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R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl, and

235

R⁴ is, independently for each compound of Formula (A8), selected from the group consisting of:

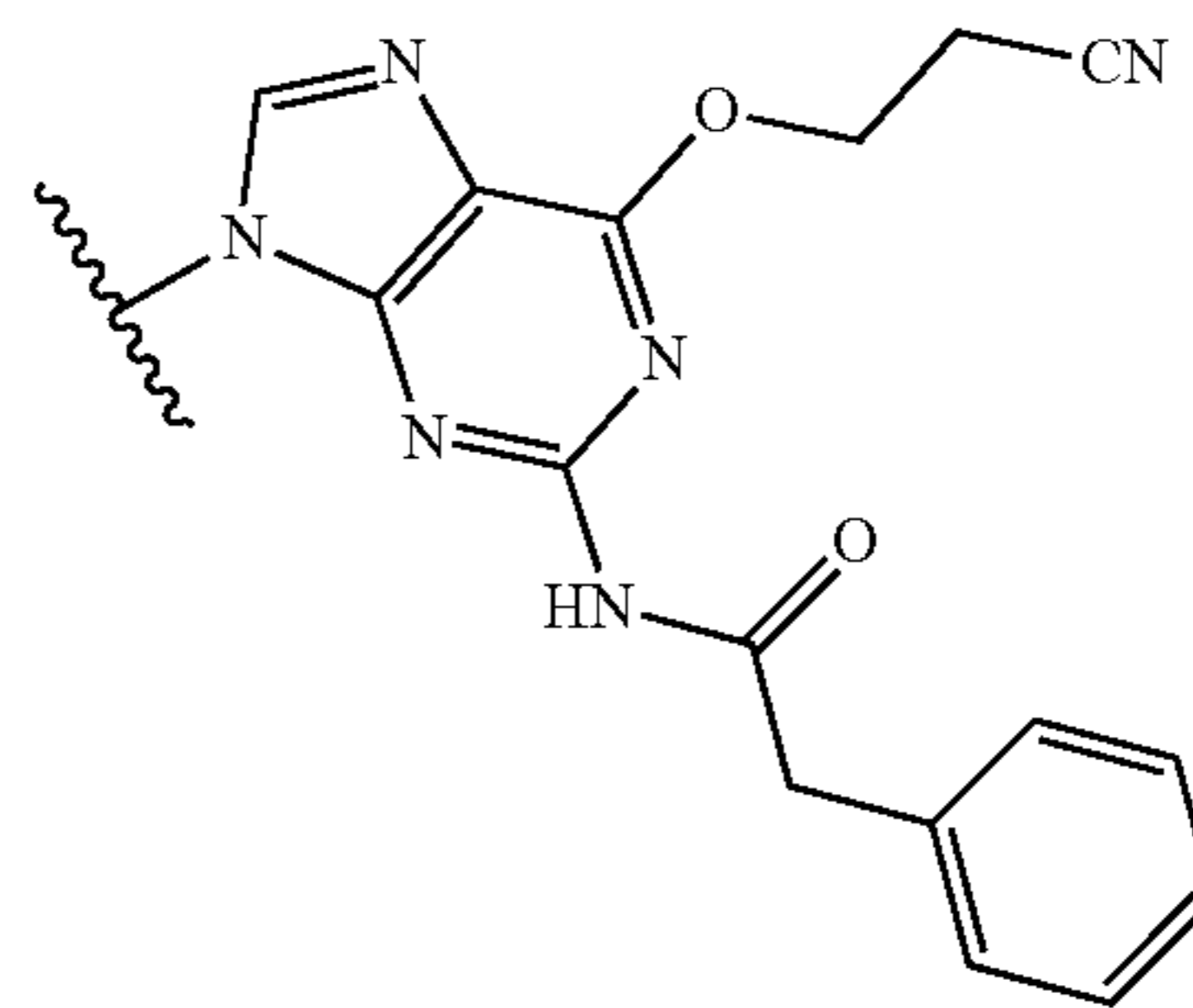


236

-continued

(PG)

(PC) 5



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(DPG)

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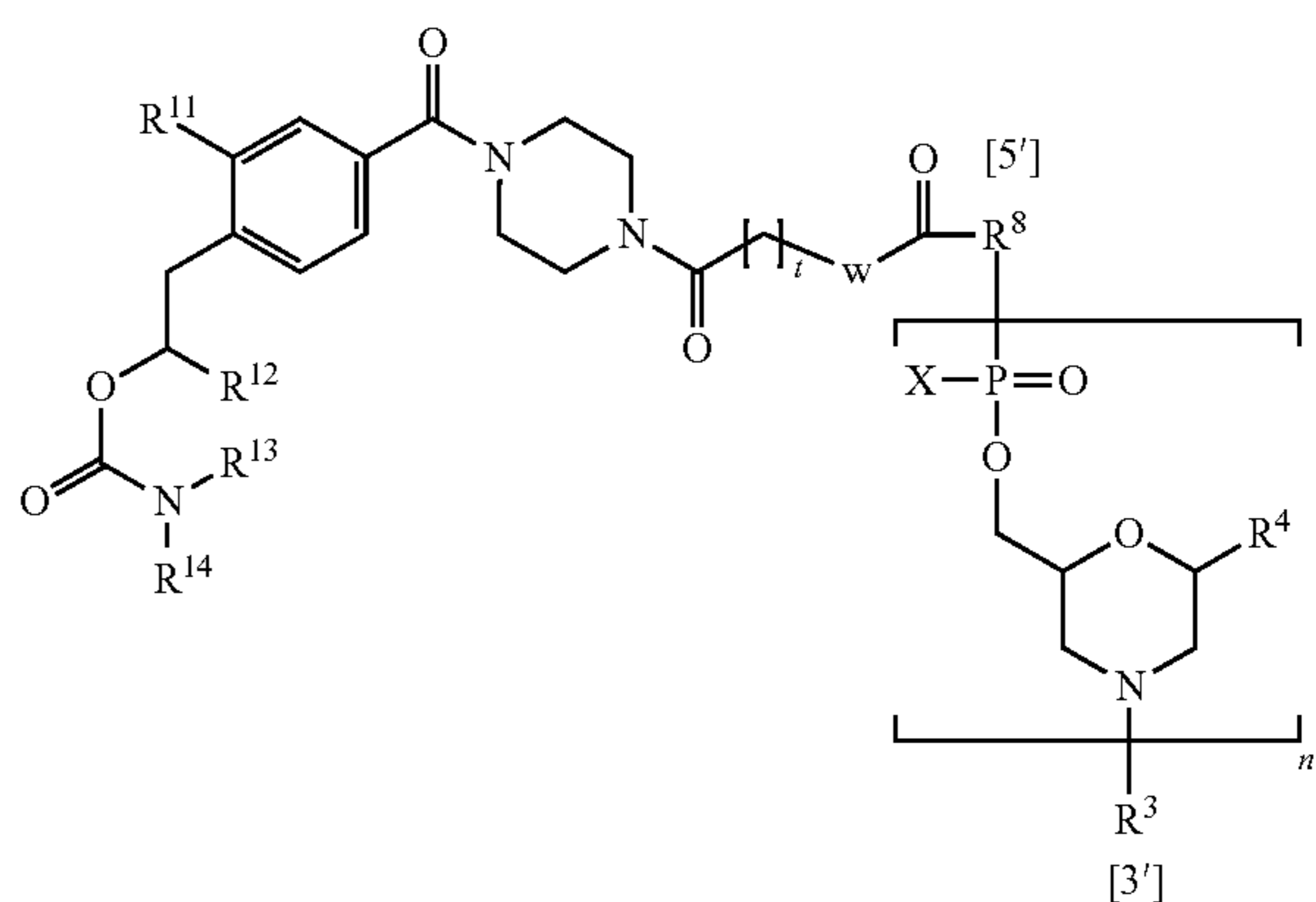
to form a compound of Formula (B9):

(B9)

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(T) 30



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wherein:

w is selected from O or C;

t is 1, 2, 3, 4, or 5;

R¹¹ is selected from the group consisting of halo, CN, and NO₂;

R¹² is C₁₋₆-alkyl;

R¹³ is H or C₁₋₆-alkyl;

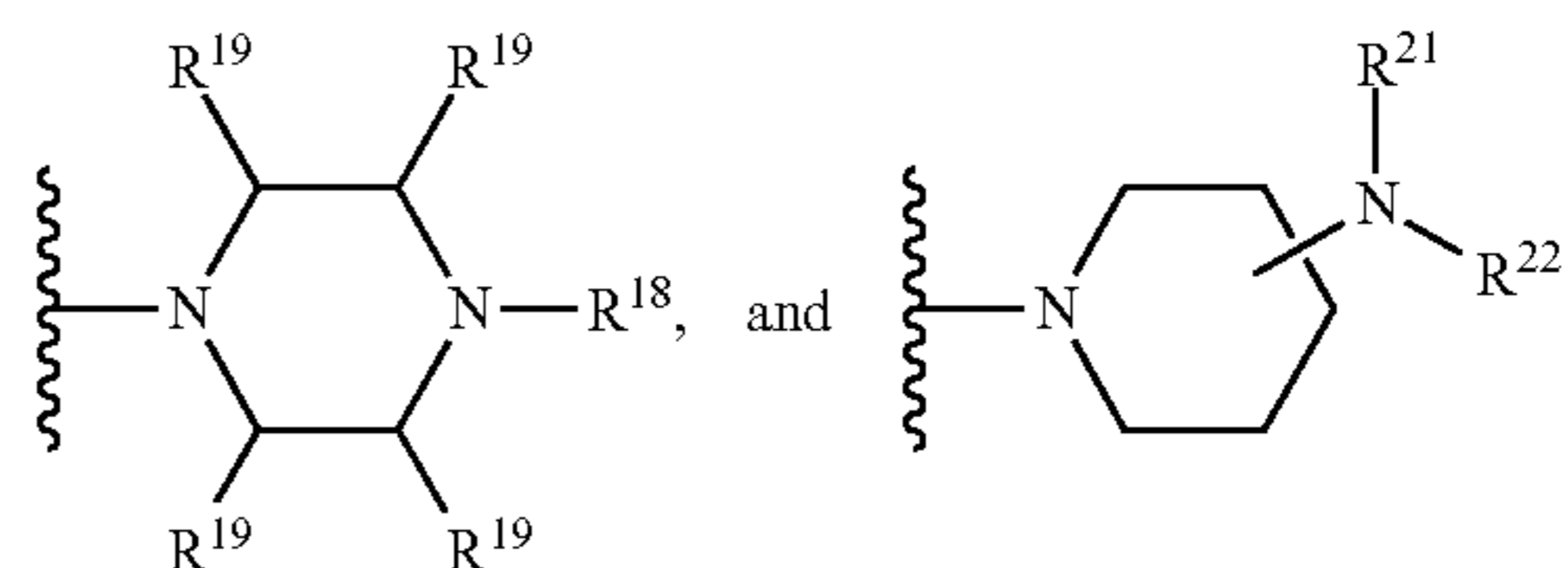
R¹⁴ is a first linker bound to a support-medium;

n is an integer from 10 to 40,

X is, independently at each occurrence, selected from H, OH, —OCH₂CH₂CN, N(R¹⁷)₂,

(P5mC) 45

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(U) 55

wherein:

R¹⁷ is, independently for each occurrence, H or C₁₋₆-alkyl;

R¹⁸ is selected from the group consisting of H, C₁₋₆-alkyl, —C(=NH)NH₂,

—C(O)(CH₂)_qNR²⁰C(=NH)NH₂,

and —C(O)(CH₂)₂NHC(O)(CH₂)_rNR²⁰C(=NH)NH₂;

q is 1, 2, 3, 4, or 5;

R¹⁹ is, independently for each occurrence, H or methyl;

R²⁰ is H or C₁₋₆-alkyl;

R²¹ is selected from the group consisting of H, C₁₋₆-alkyl, C(=NH)NH₂, C(O)(CH₂)_rNR²³C(=NH)

(I) 65

237

NH_2 , $\text{C(O)CH(NH}_2\text{)(CH}_2\text{)}_3\text{NHC(=NH)NH}_2$, $\text{C(O)(CH}_2\text{)}_2\text{NHC(O)(CH}_2\text{)}_5\text{NR}^{23}\text{C(=NH)NH}_2$, and $\text{C(O)CH(NH}_2\text{)(CH}_2\text{)}_4\text{NH}_2$;

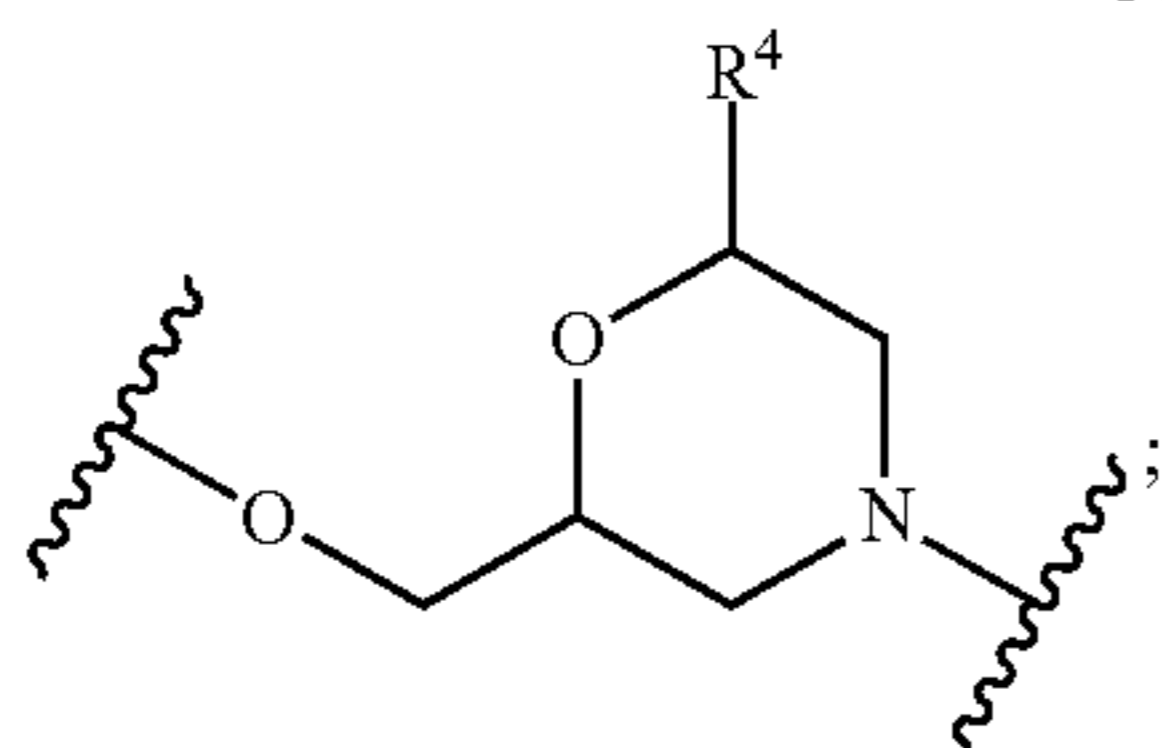
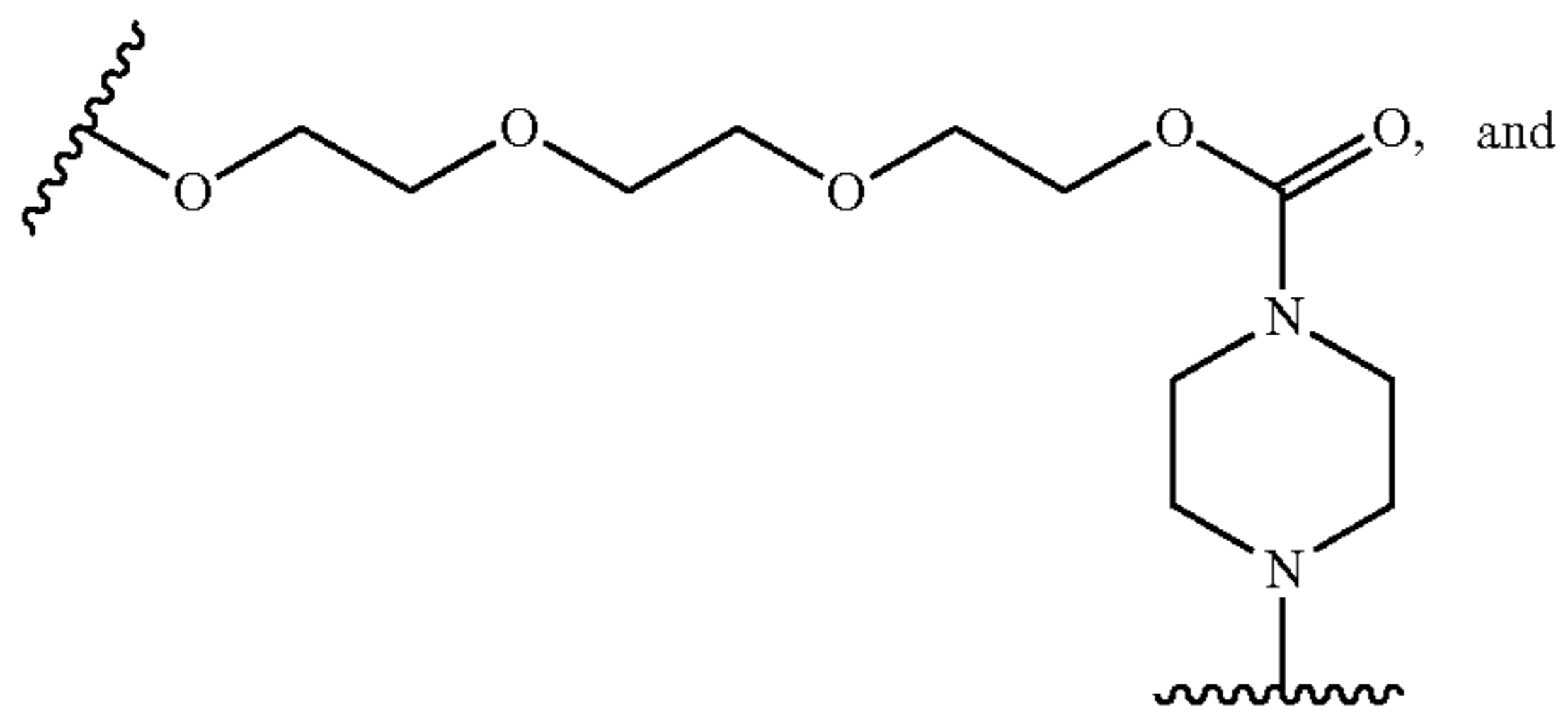
R^{22} is H or C_{1-6} -alkyl;

R^{23} is H or C_{1-6} -alkyl; and

r is 1, 2, 3, 4, or 5;

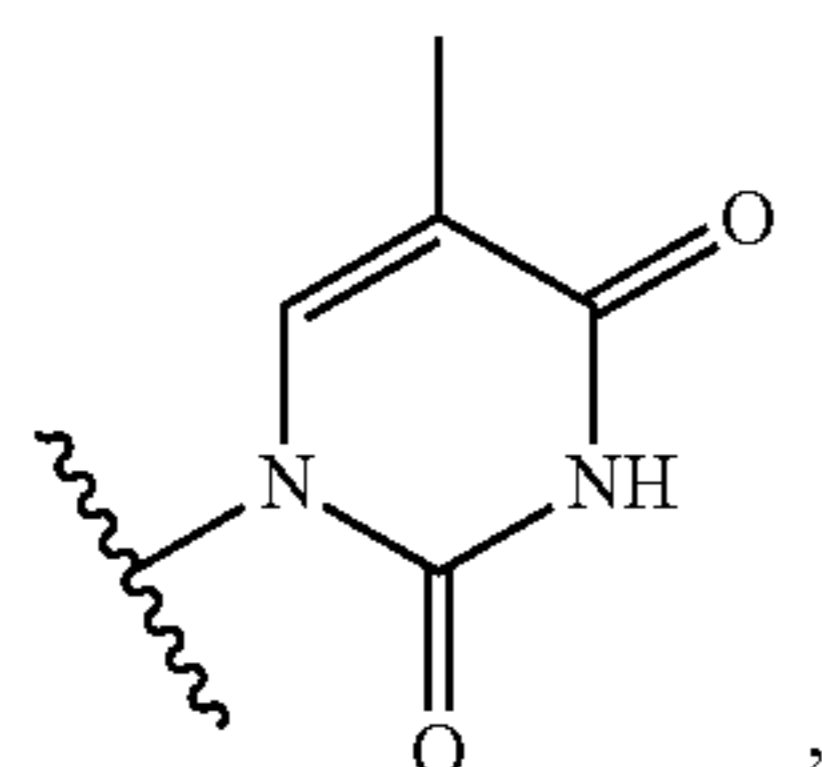
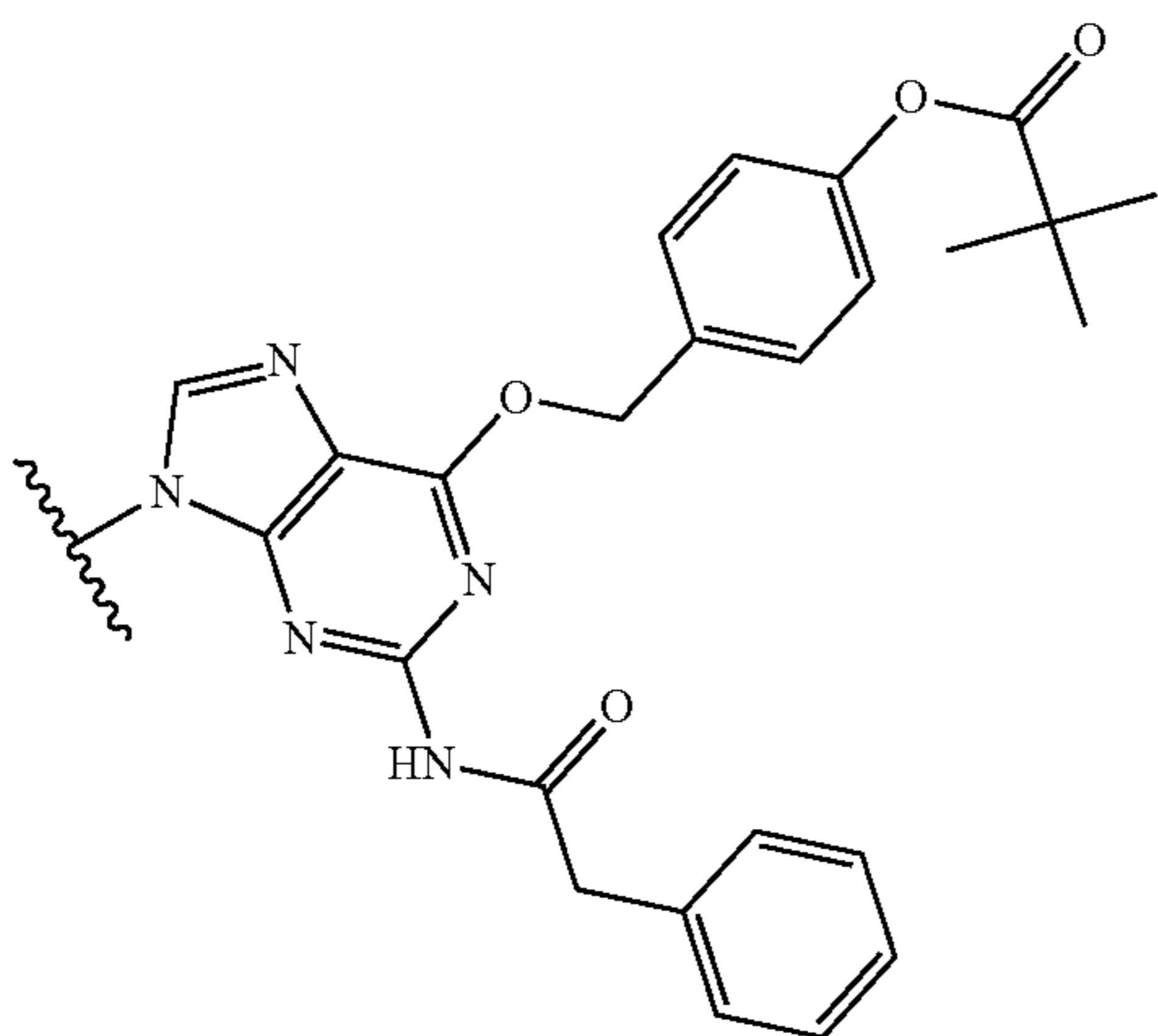
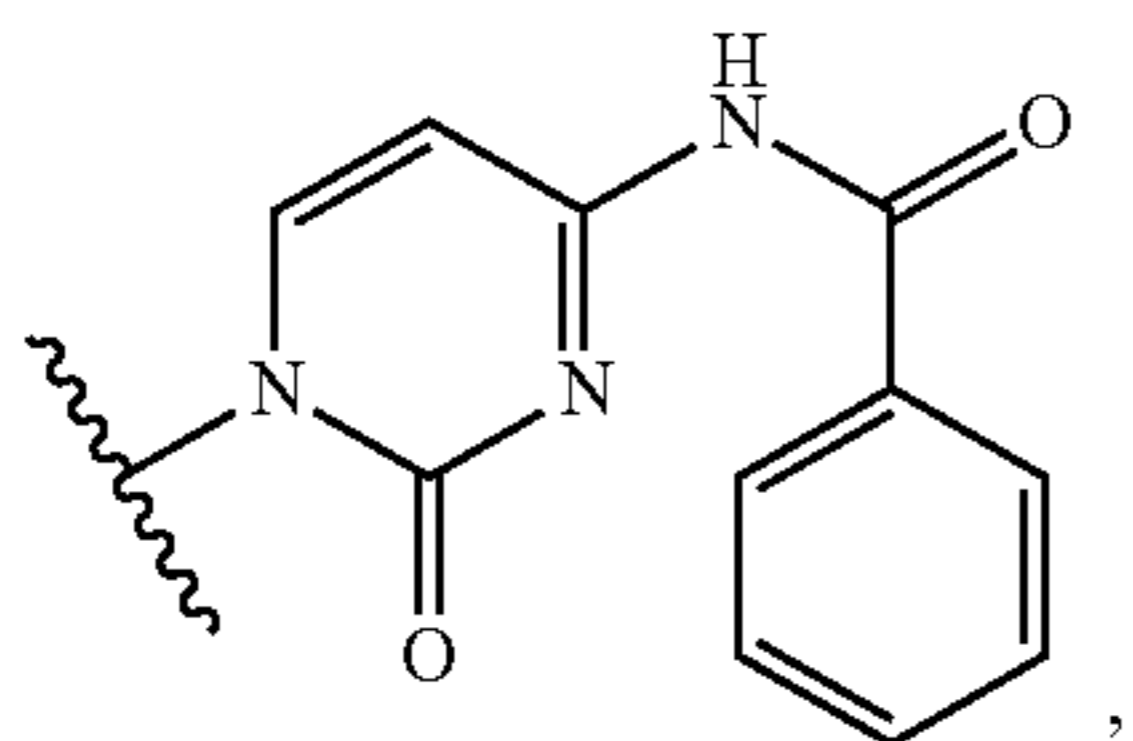
R^3 is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl,

R^8 is selected from:



and

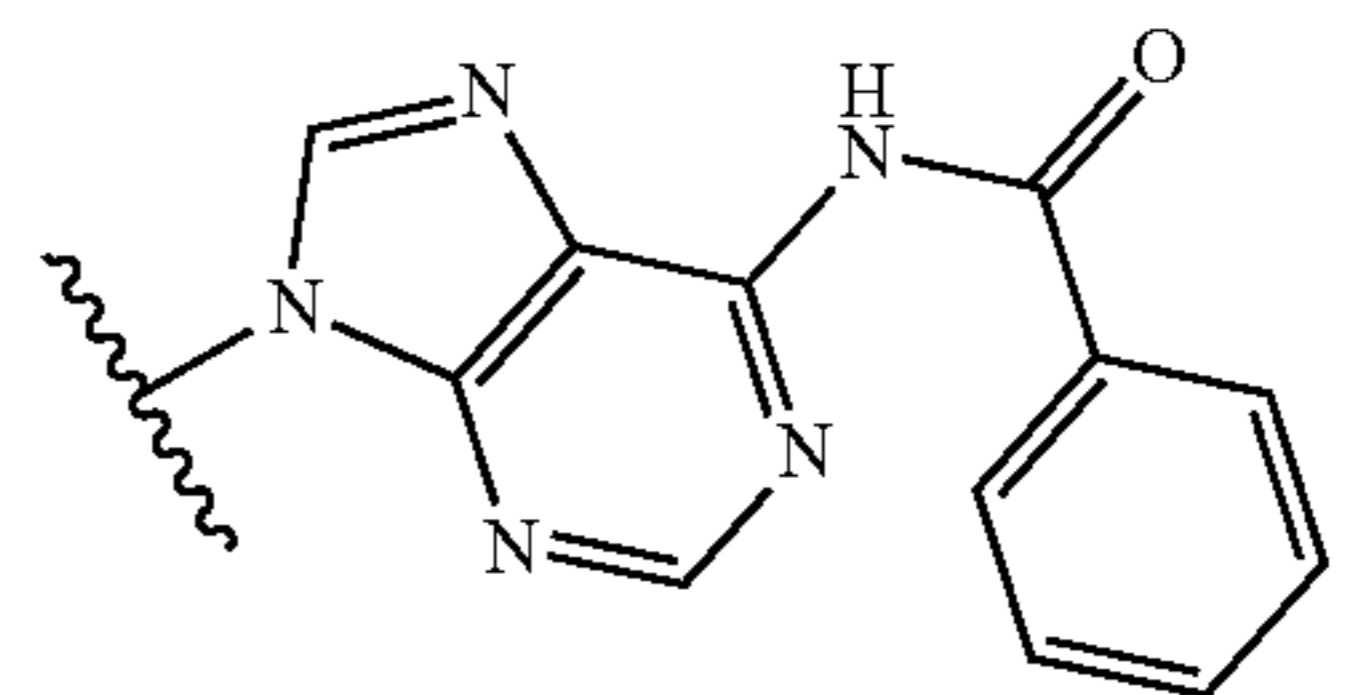
R^4 is, independently for each occurrence, selected from the group consisting of:



238

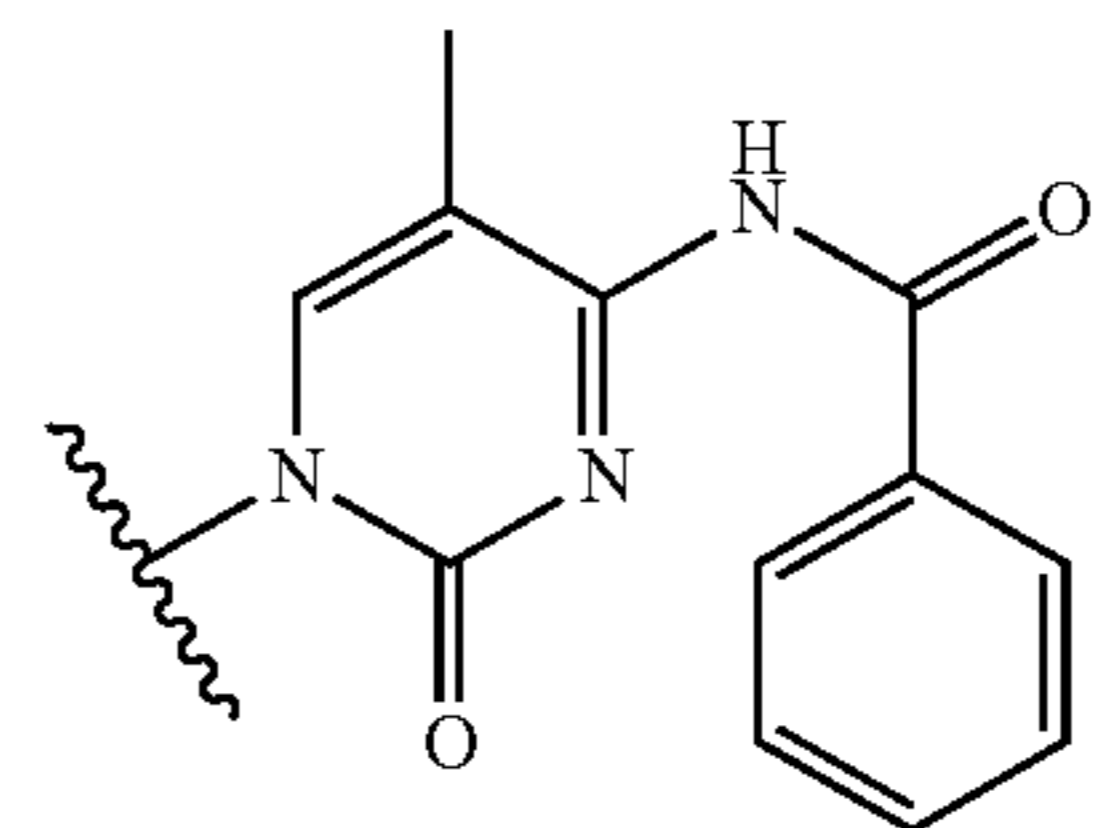
-continued

(PA)



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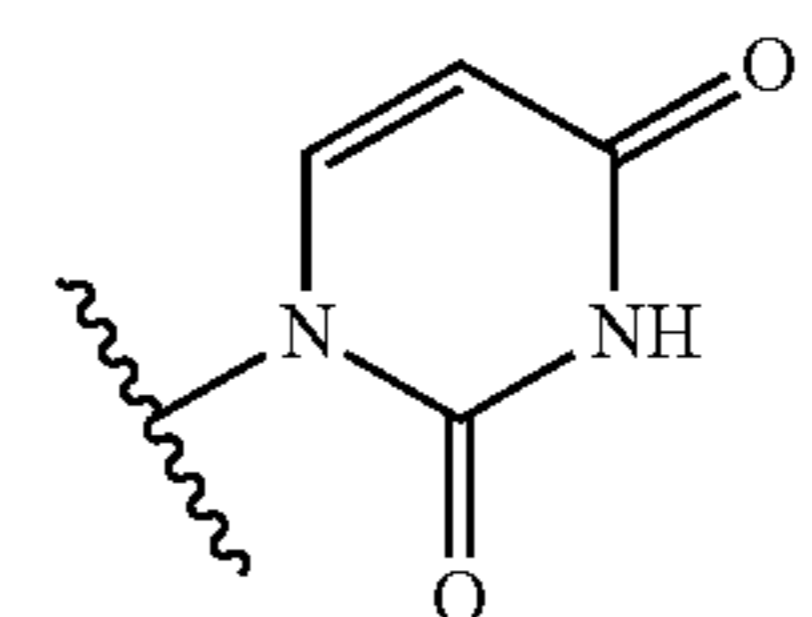
(P5mC)



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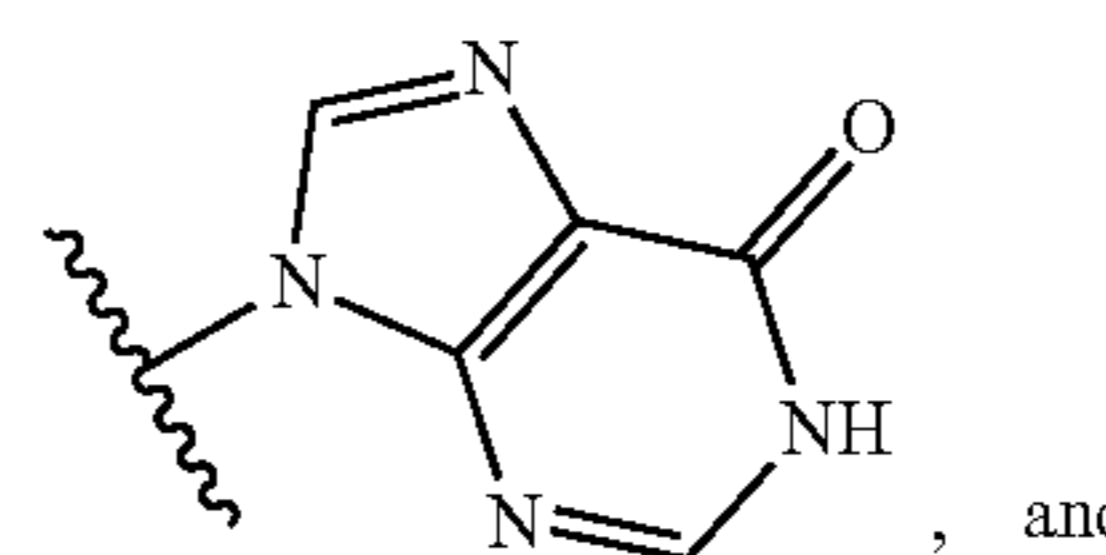
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(U)



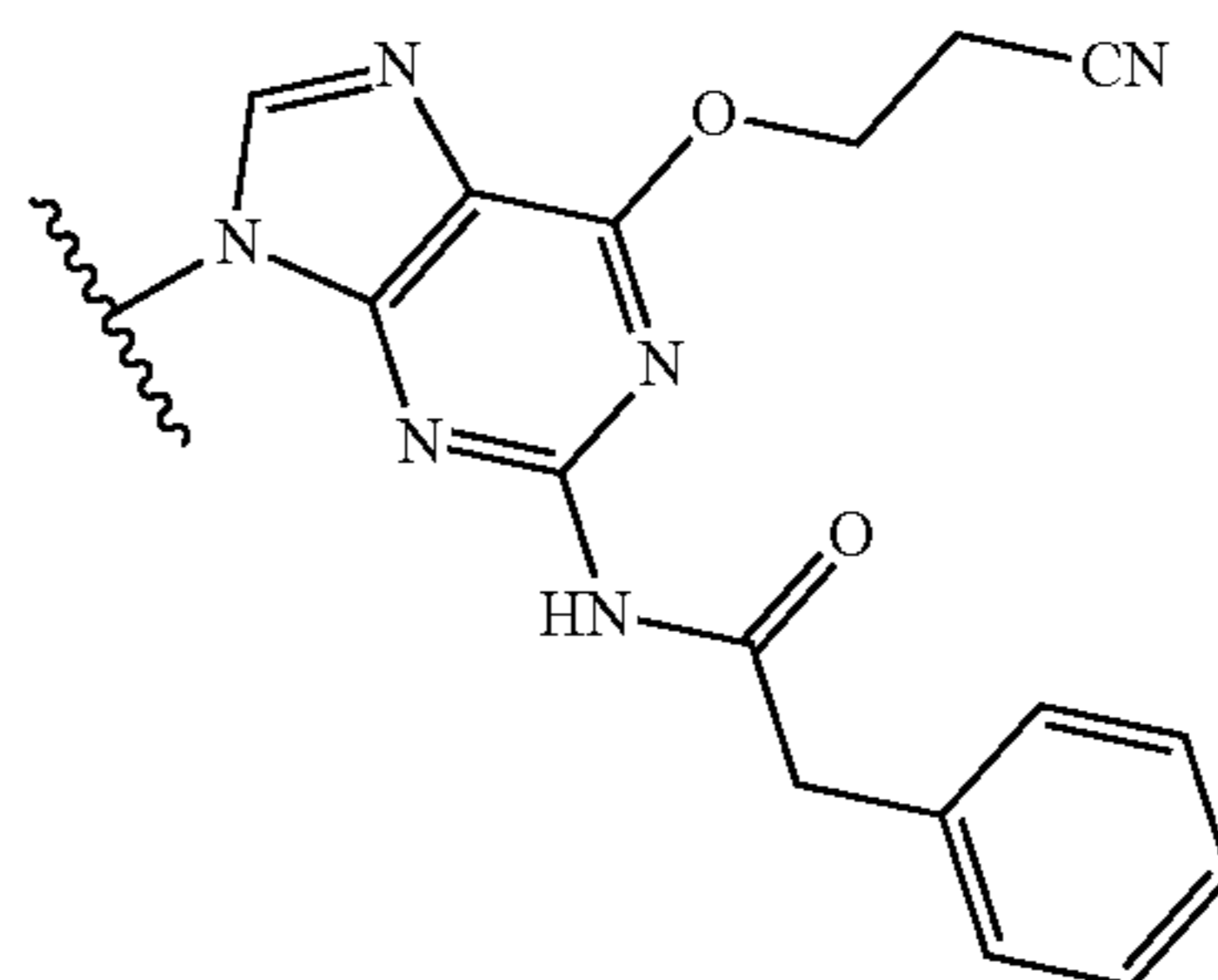
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(I)



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(PG)



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(PC)

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(DPG)

and

(f) contacting the compound of Formula (A9) with a deblocking agent to form a compound of Formula (B10):

(B10)

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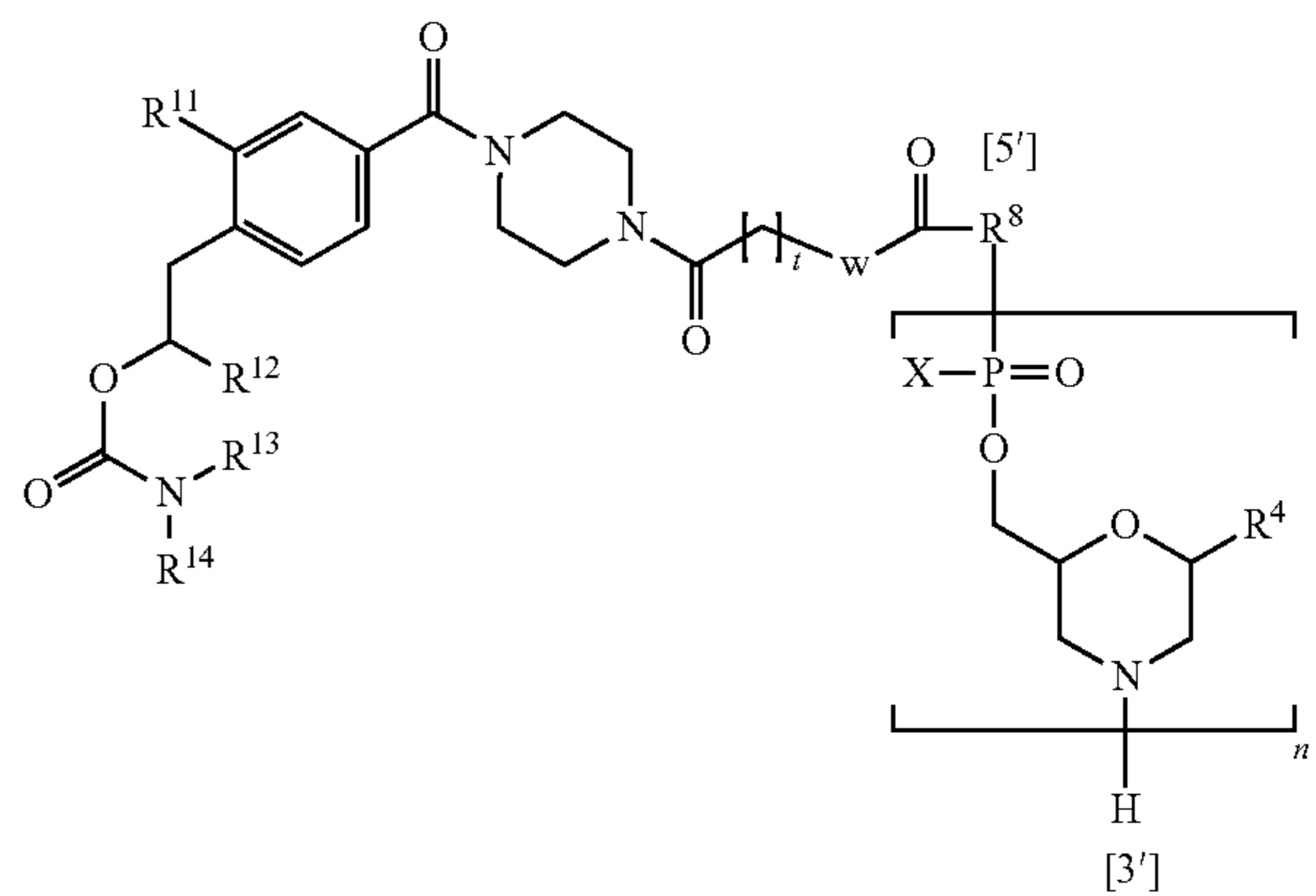
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(T)

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wherein:

w is selected from 0 or C;

t is 1, 2, 3, 4, or 5;

239

R¹¹ is selected from the group consisting of halo, CN, and NO₂;

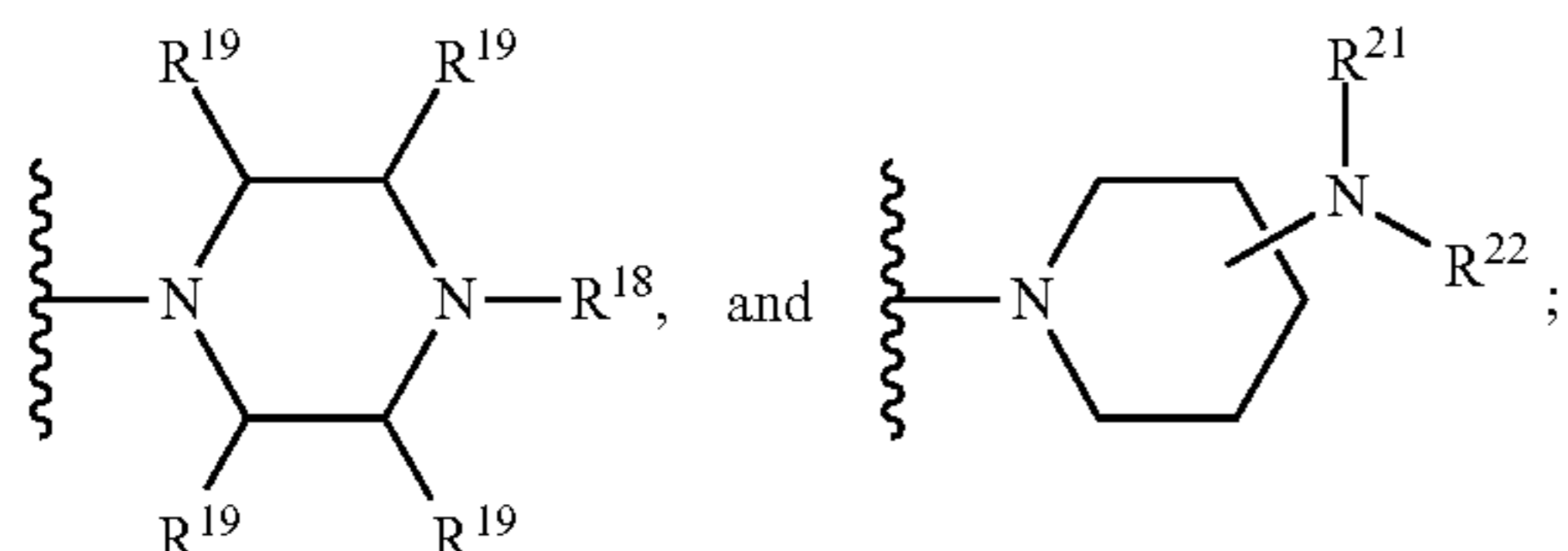
R¹² is C₁₋₆-alkyl;

R¹³ is H or C₁₋₆-alkyl;

R¹⁴ is a first linker bound to a support-medium;

n is an integer from 10 to 40,

X is, independently at each occurrence, selected from H, OH, —OCH₂CH₂CN, N(R¹⁷)₂,



wherein:

R¹⁷ is, independently for each occurrence, H or C₁₋₆-alkyl;

R¹⁸ is selected from the group consisting of H, C₁₋₆-alkyl, —C(=NH)NH₂,

—C(O)(CH₂)_qNR²⁰C(=NH)NH₂,

and —C(O)(CH₂)₂NHC(O)(CH₂)₅NR²⁰C(=NH)NH₂;

q is 1, 2, 3, 4, or 5;

R¹⁹ is, independently for each occurrence, H or methyl;

R²⁰ is H or C₁₋₆-alkyl;

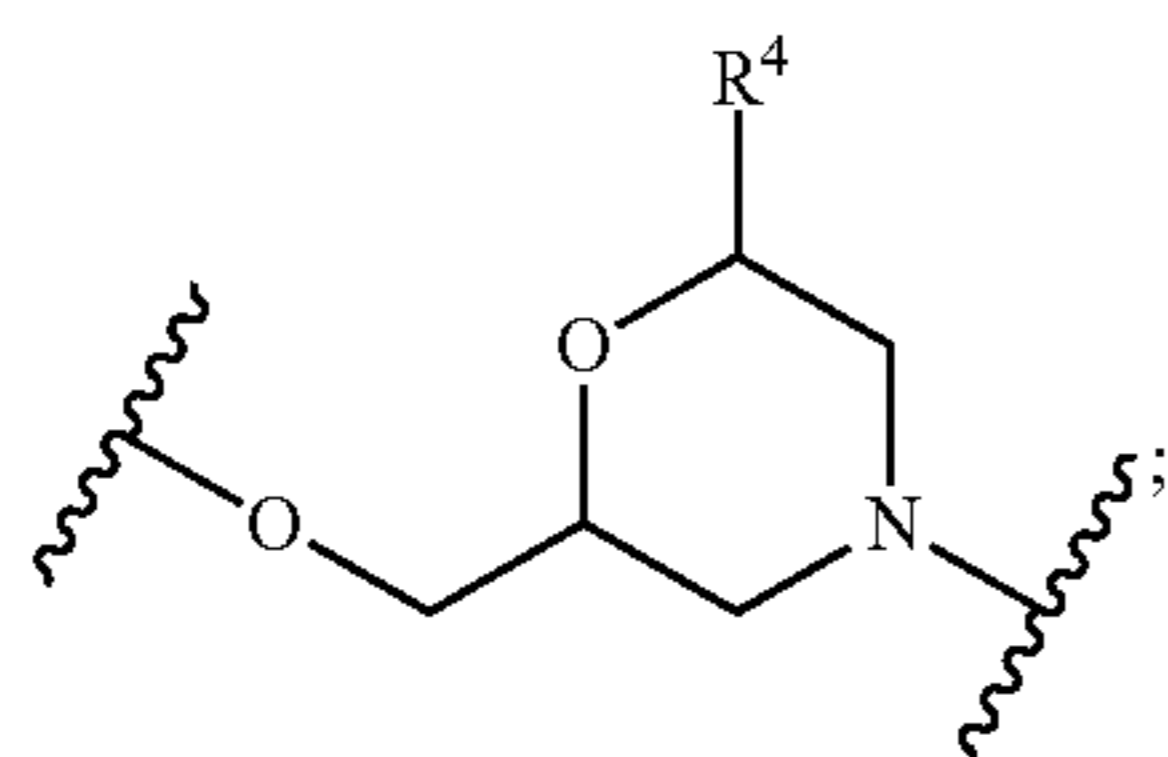
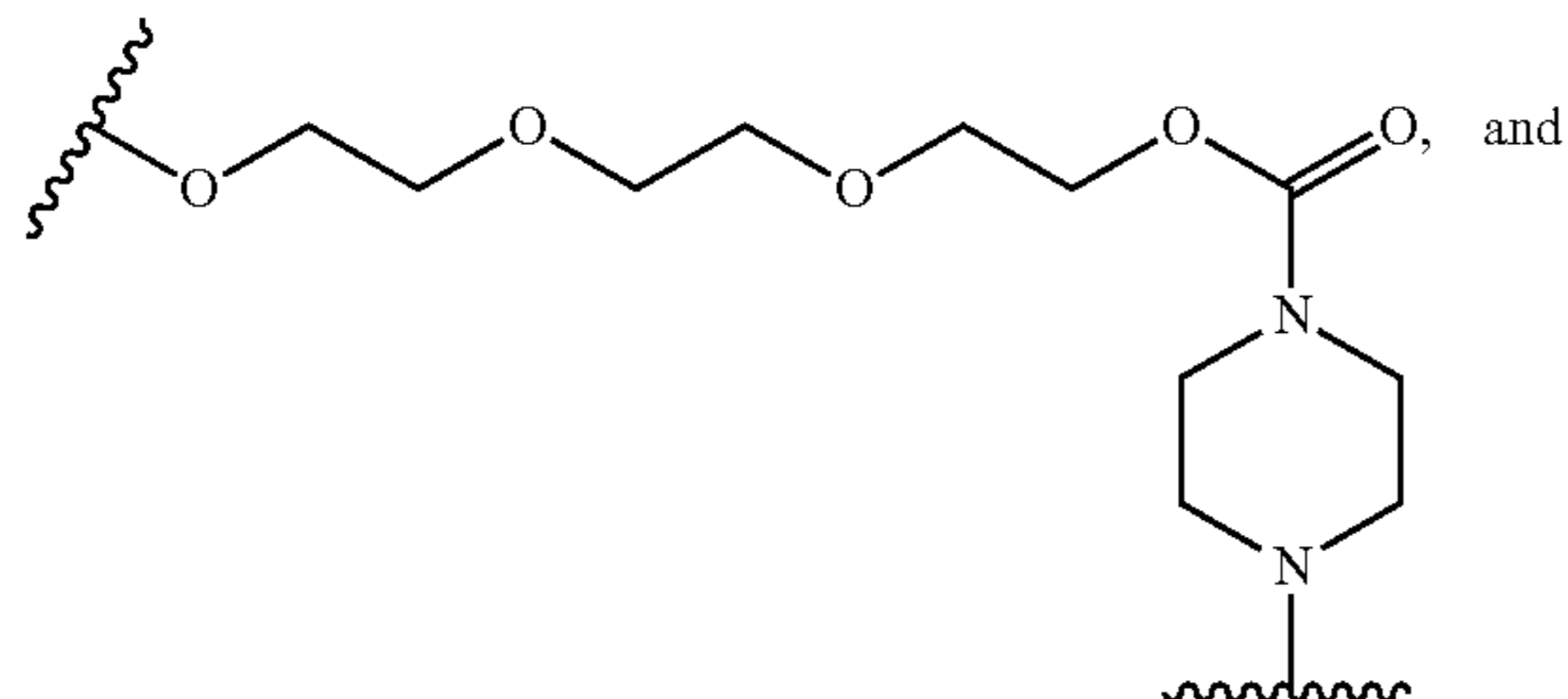
R²¹ is selected from the group consisting of H, C₁₋₆-alkyl, C(=NH)NH₂, C(O)(CH₂)_rNR²³C(=NH)NH₂, C(O)CH(NH₂)(CH₂)₃NHC(=NH)NH₂, C(O)(CH₂)₂NHC(O)(CH₂)₅NR²³C(=NH)NH₂, and C(O)CH(NH₂)(CH₂)₄NH₂;

R²² is H or C₁₋₆-alkyl;

R²³ is H or C₁₋₆-alkyl; and

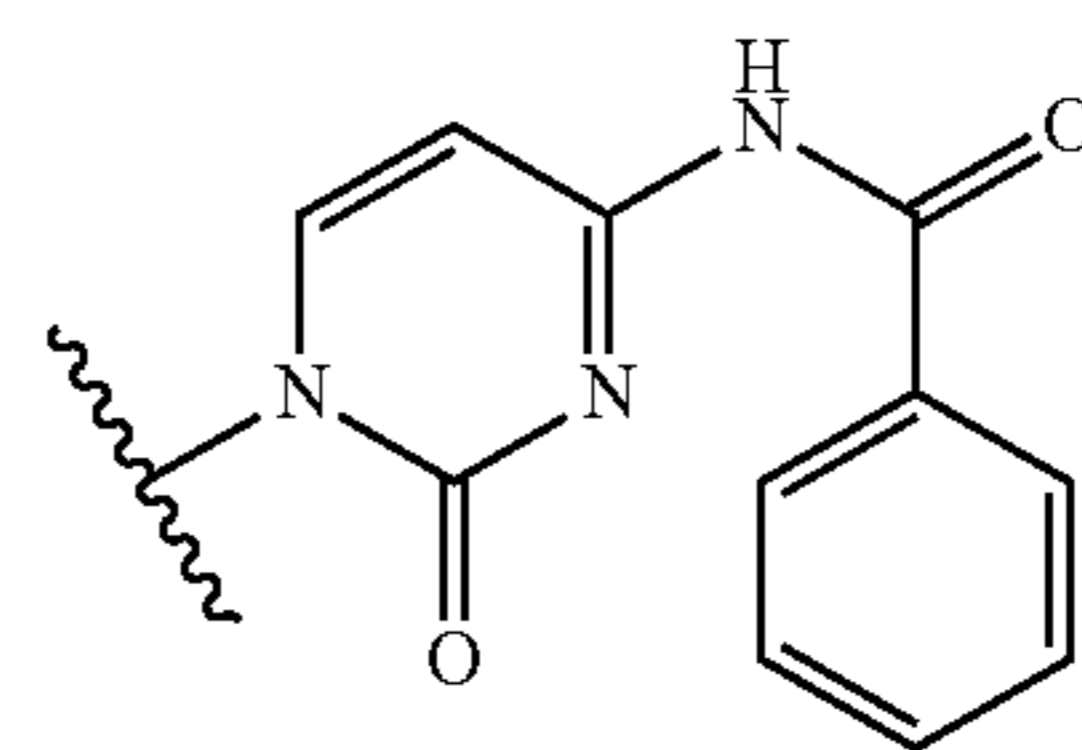
r is 1, 2, 3, 4, or 5;

R⁸ is selected from:

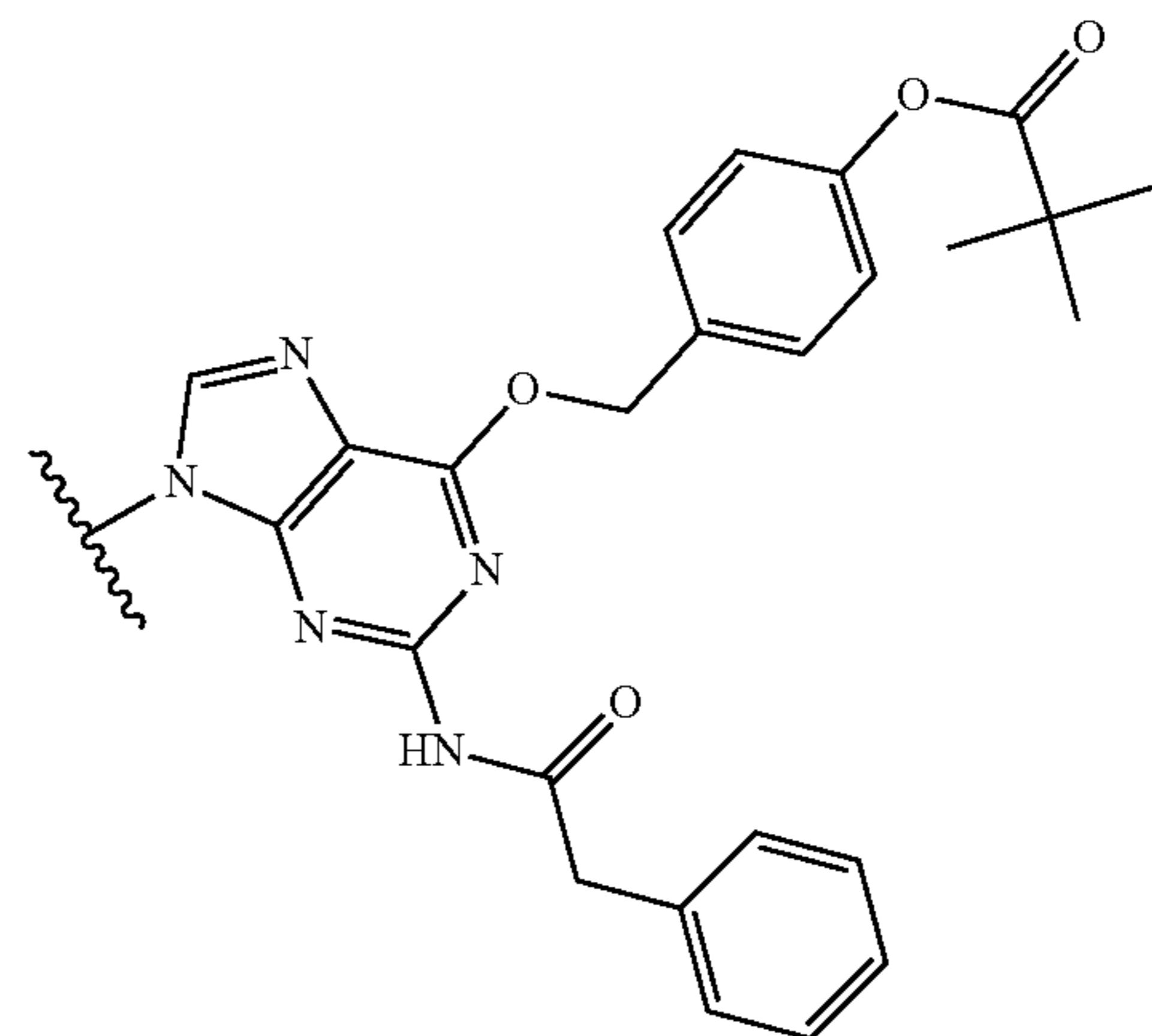


R⁴ is, independently for each occurrence, selected from the group consisting of:

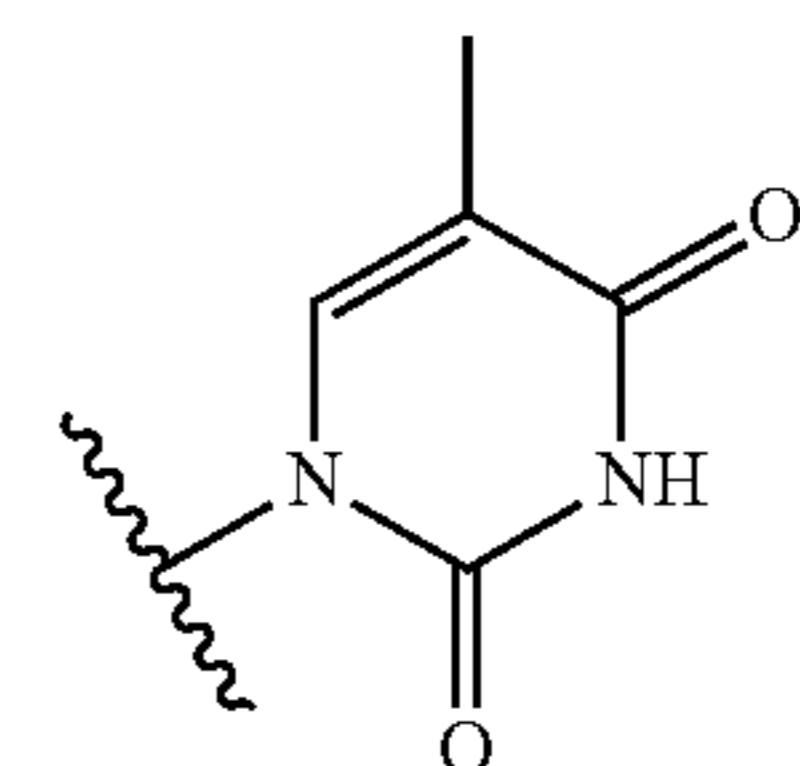
240



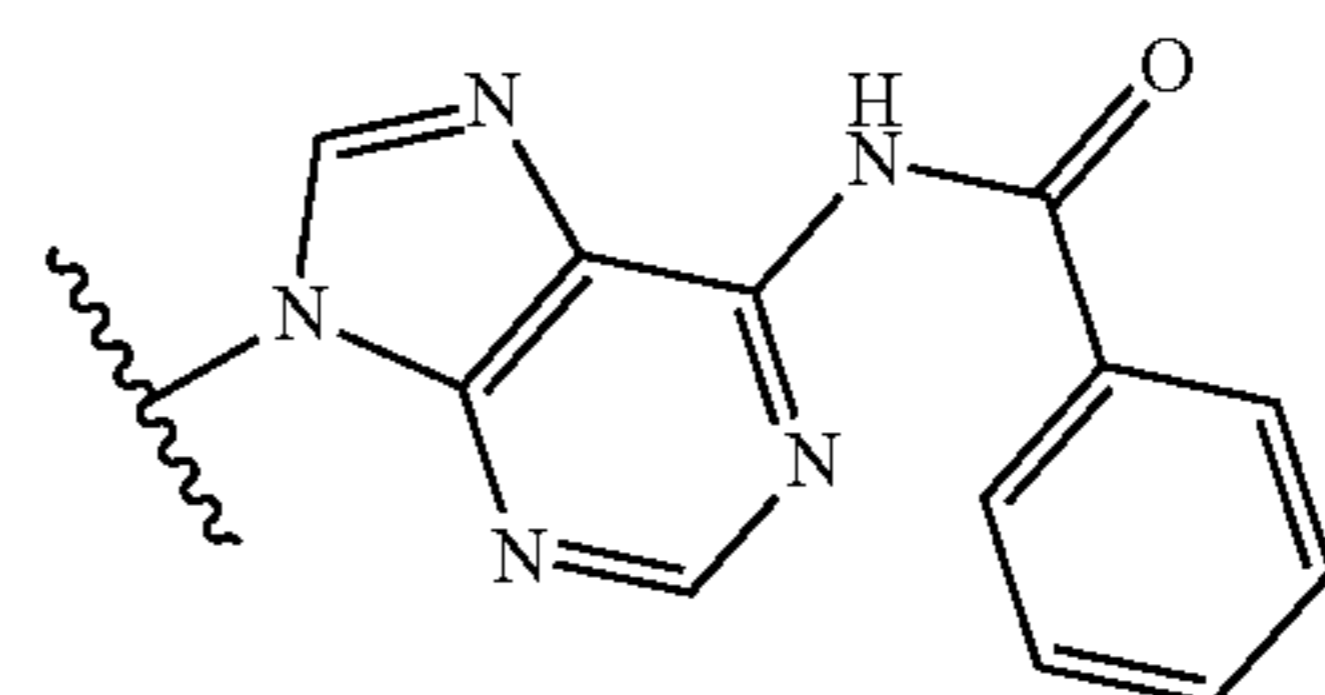
(PC)



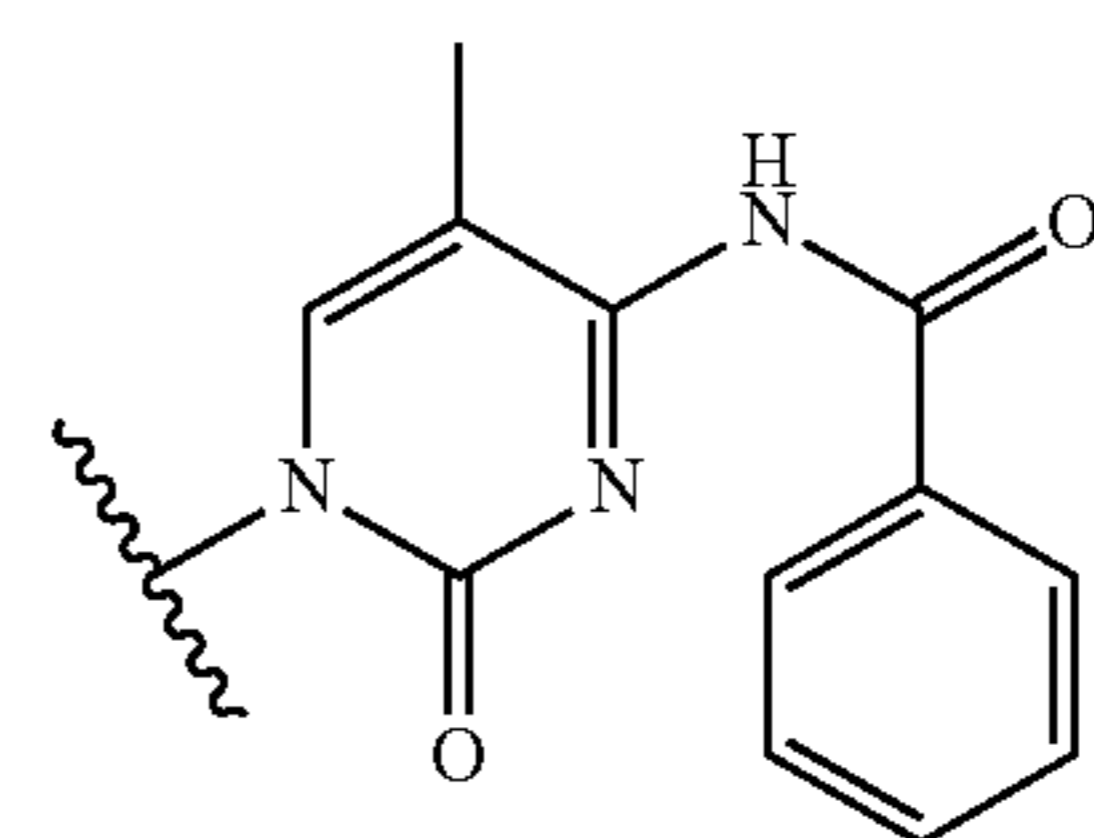
(DPG)



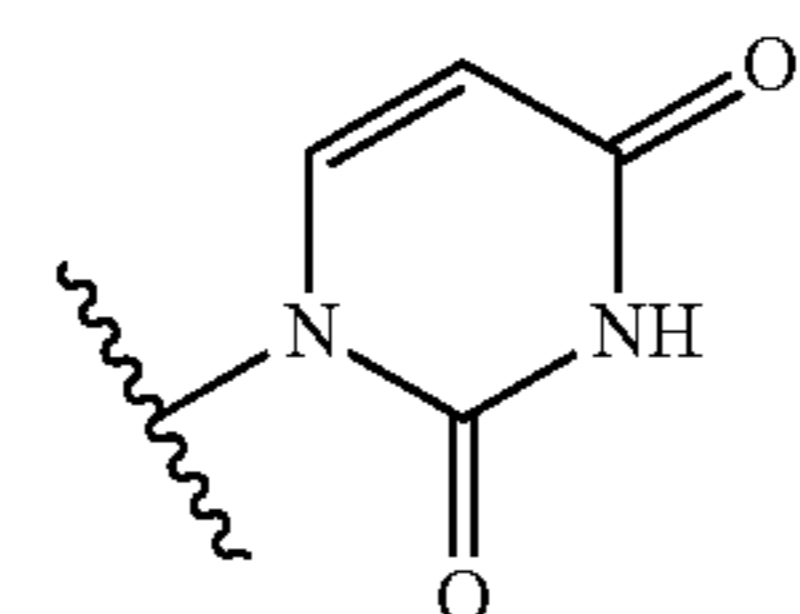
(T)



(PA)



(P5mC)



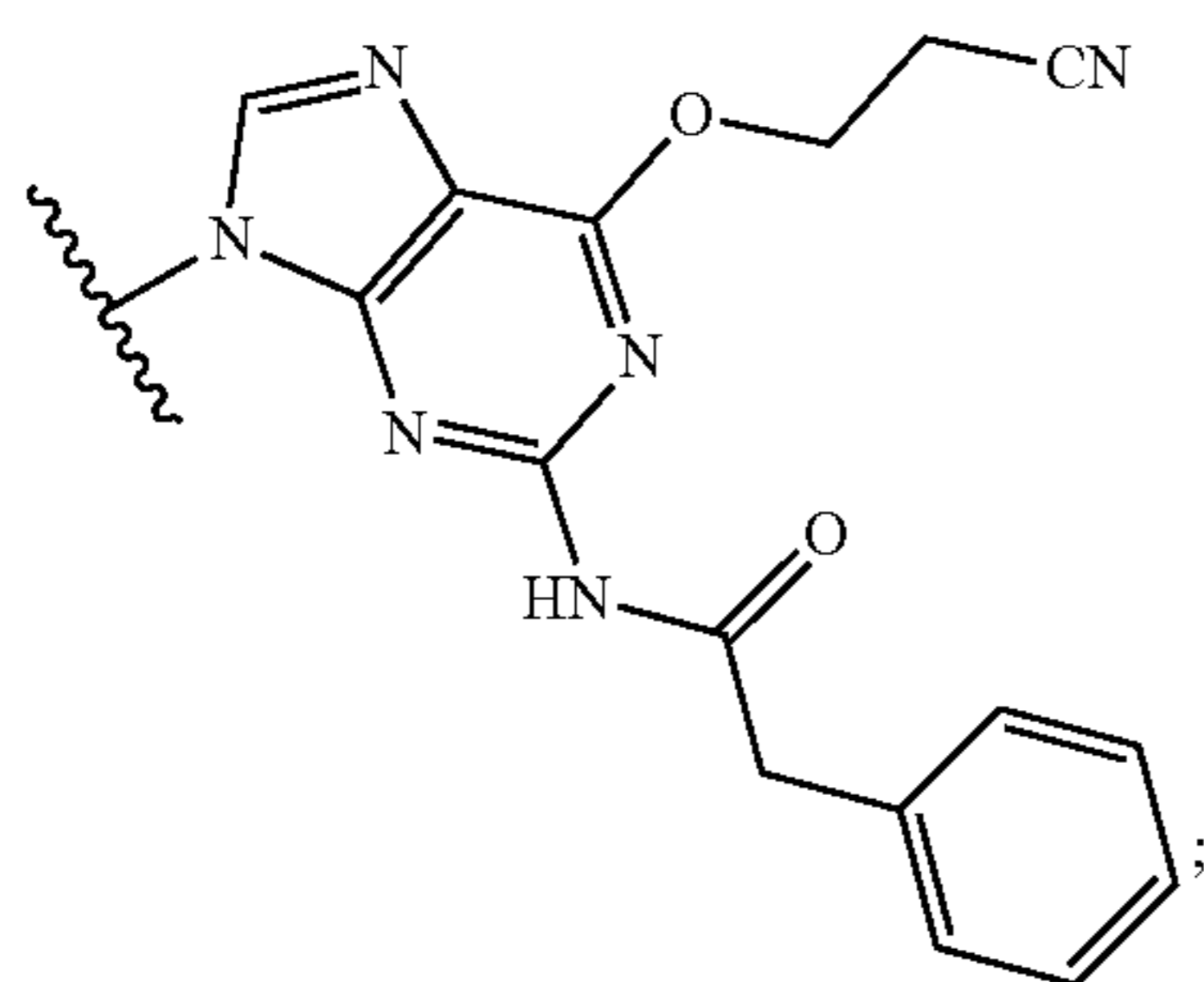
(U)



(I)

241

-continued

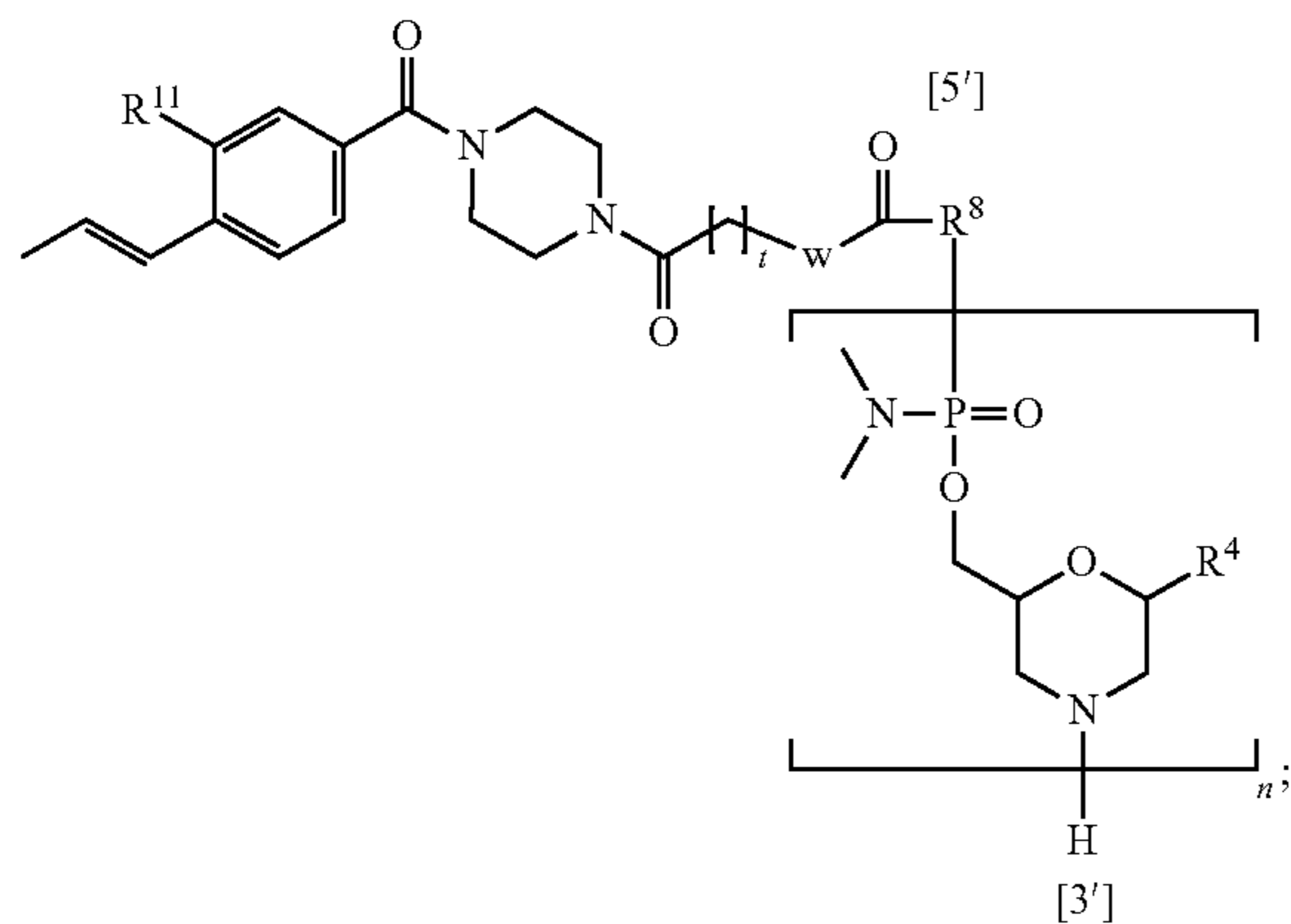


(PG)

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(g) contacting the compound of Formula (B10) with a cleaving agent to form a compound of Formula (B11):



(B11)

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30 and

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wherein:

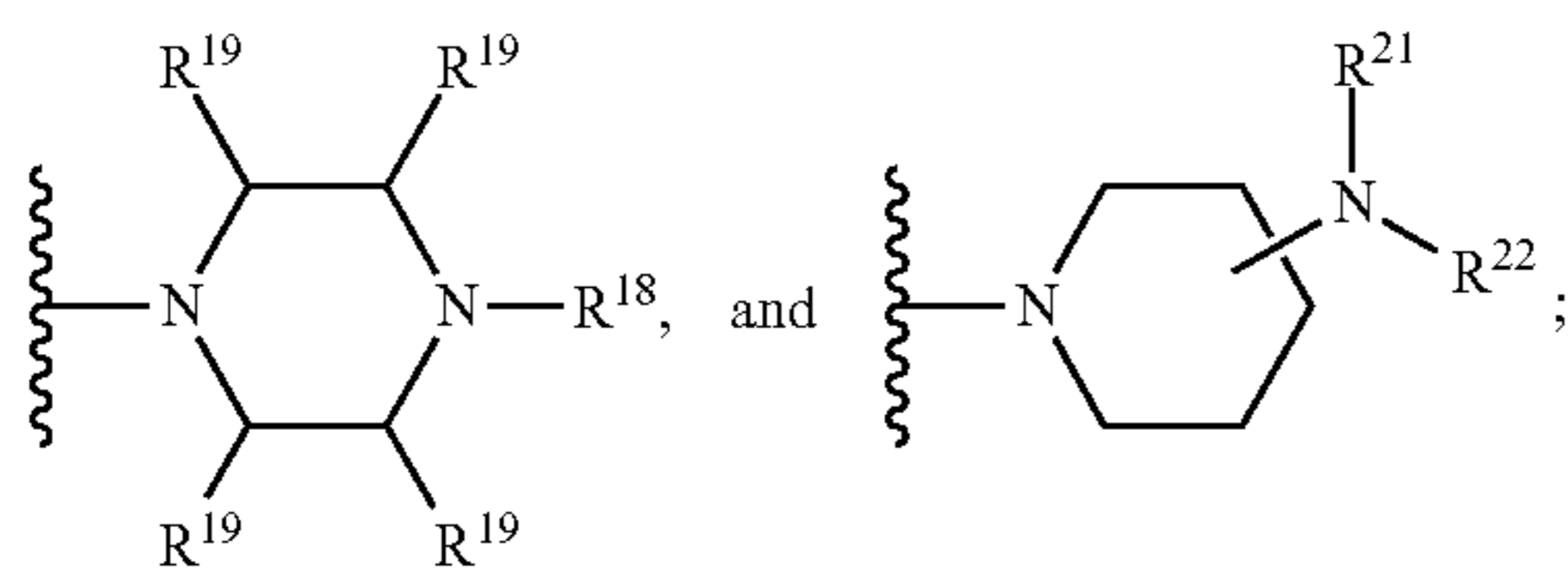
w is selected from O or C;

t is 1, 2, 3, 4, or 5;

R¹¹ is selected from the group consisting of halo, CN, and NO₂;

n is an integer from 10 to 40,

X is, independently at each occurrence, selected from H, OH, —OCH₂CH₂CN, N(R¹⁷)₂,



wherein:

R¹⁷ is, independently for each occurrence, H or C₁₋₆-alkyl;

R¹⁸ is selected from the group consisting of H, C₁₋₆-alkyl, —C(=NH)NH₂,

—C(O)(CH₂)_qNR²⁰C(=NH)NH₂, and —C(O)(CH₂)₂NHC(O)(CH₂)NR²⁰C(=NH)NH₂;

q is 1, 2, 3, 4, or 5;

R¹⁹ is, independently for each occurrence, H or methyl;

R²⁰ is H or C₁₋₆-alkyl;

R²¹ is selected from the group consisting of H, C₁₋₆-alkyl, C(=NH)NH₂, C(O)(CH₂)_rNR²³C(=NH)

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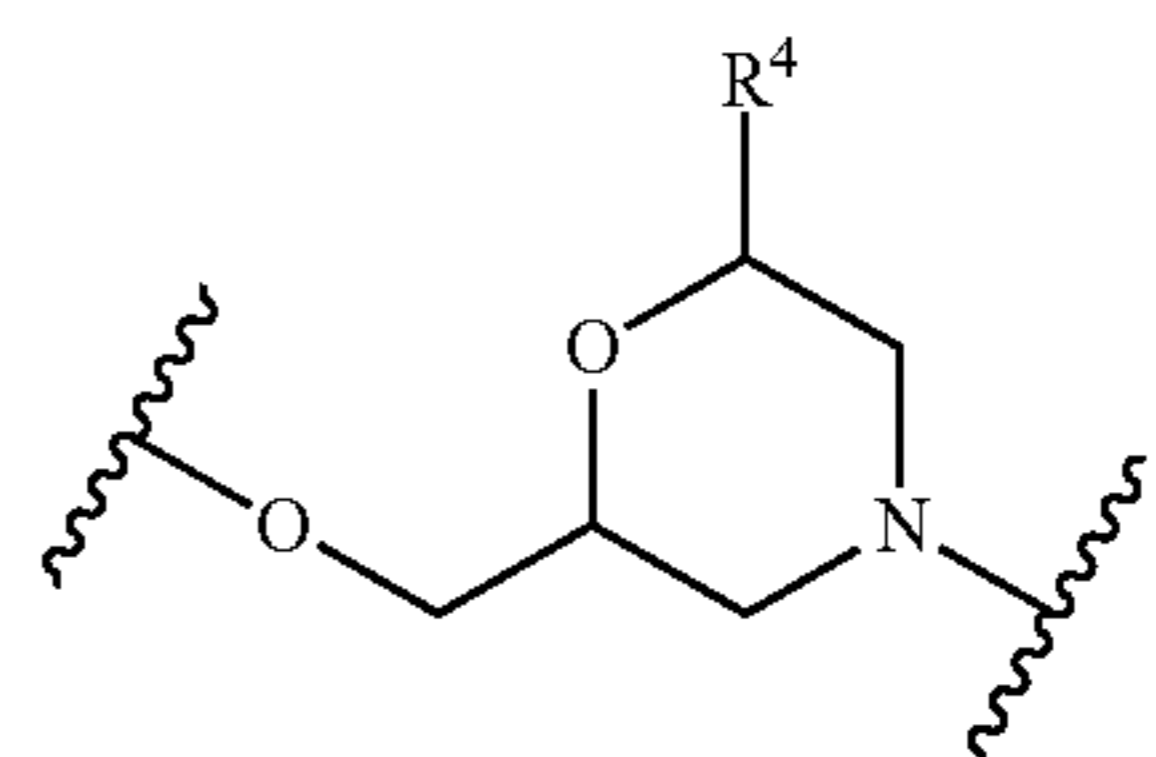
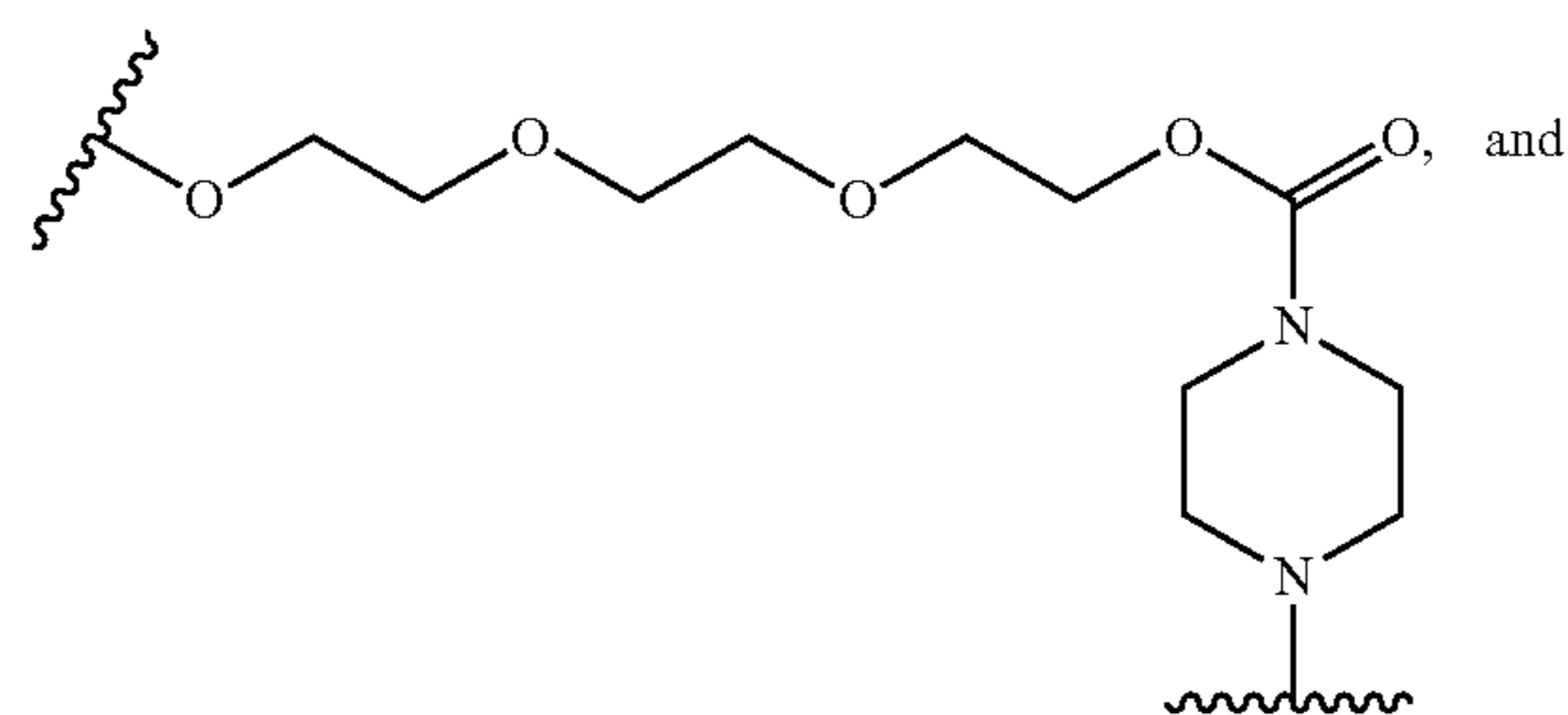
NH₂, C(O)CH(NH₂)(CH₂)₃NHC(=NH)NH₂, C(O)(CH₂)₂NHC(O)(CH₂)₅NR²³C(=NH)NH₂, and C(O)CH(NH₂)(CH₂)₄NH₂;

R²² is H or C₁₋₆-alkyl;

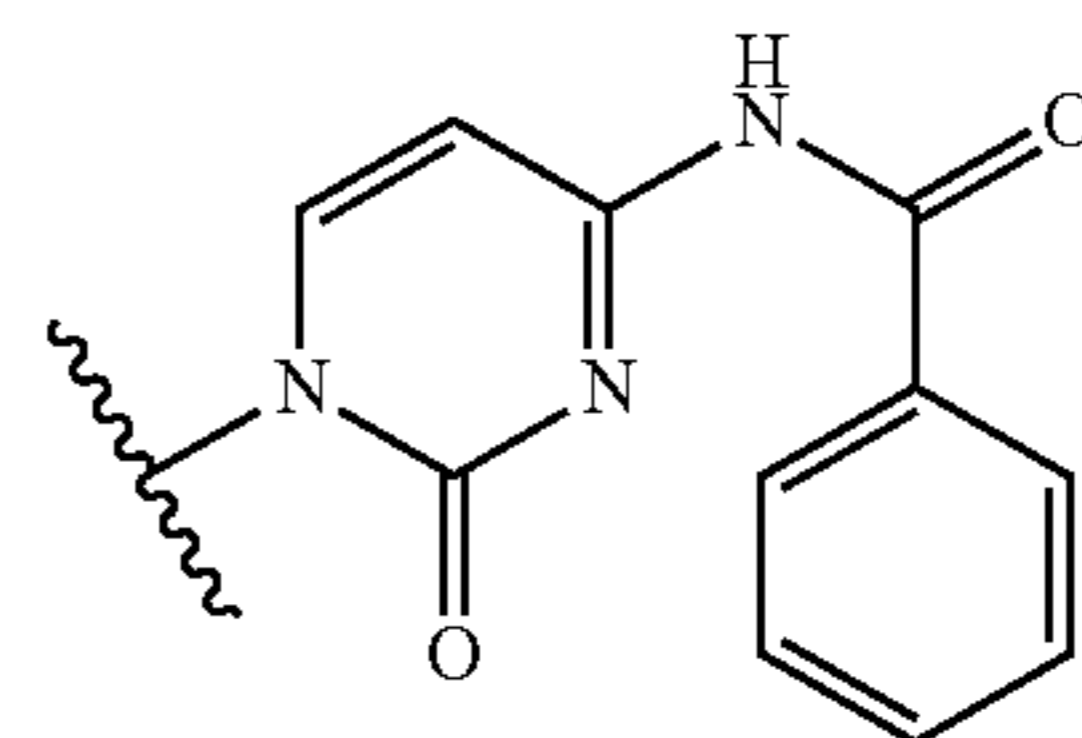
R²³ is H or C₁₋₆-alkyl; and

r is 1, 2, 3, 4, or 5;

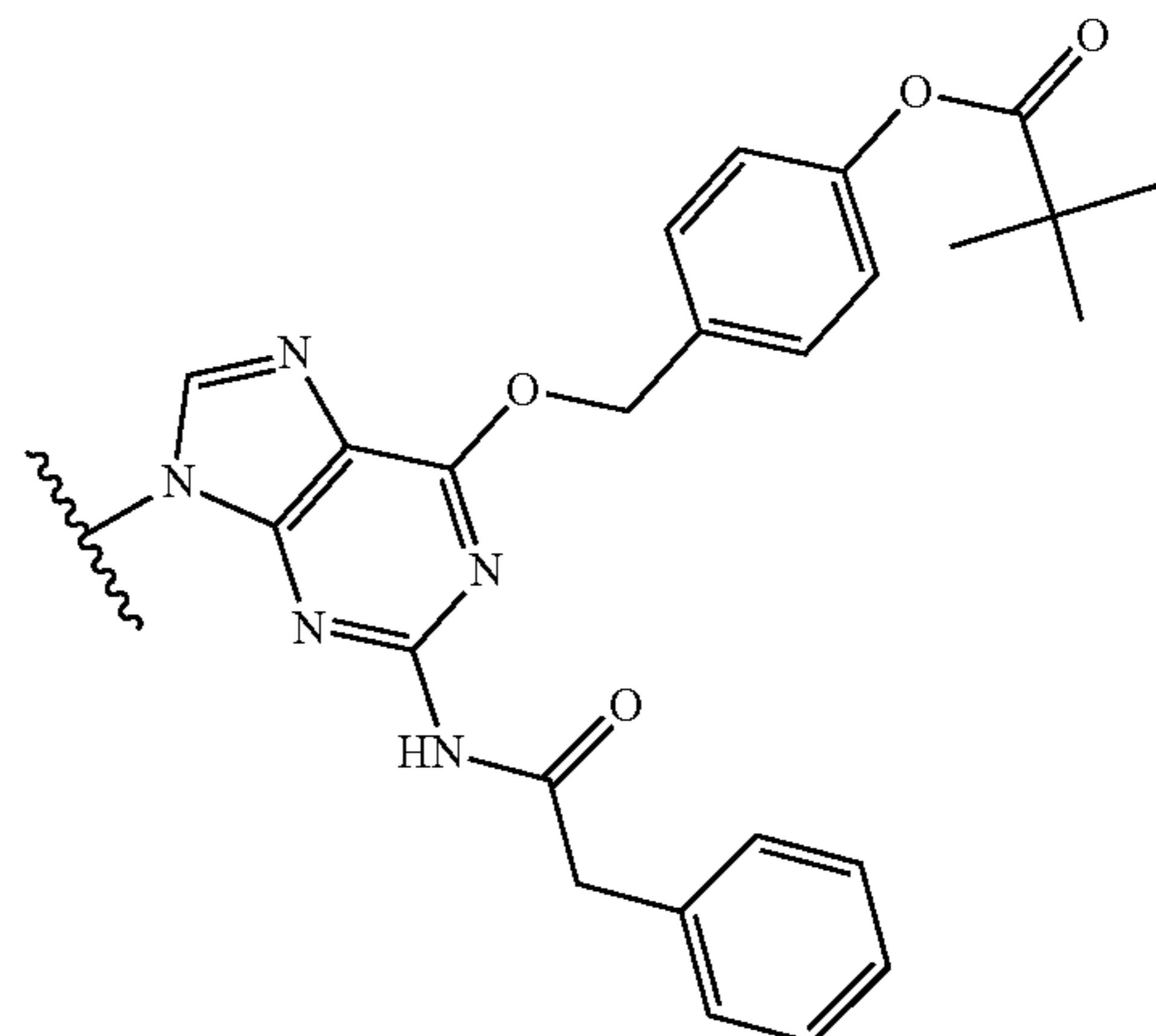
R⁸ is selected from:



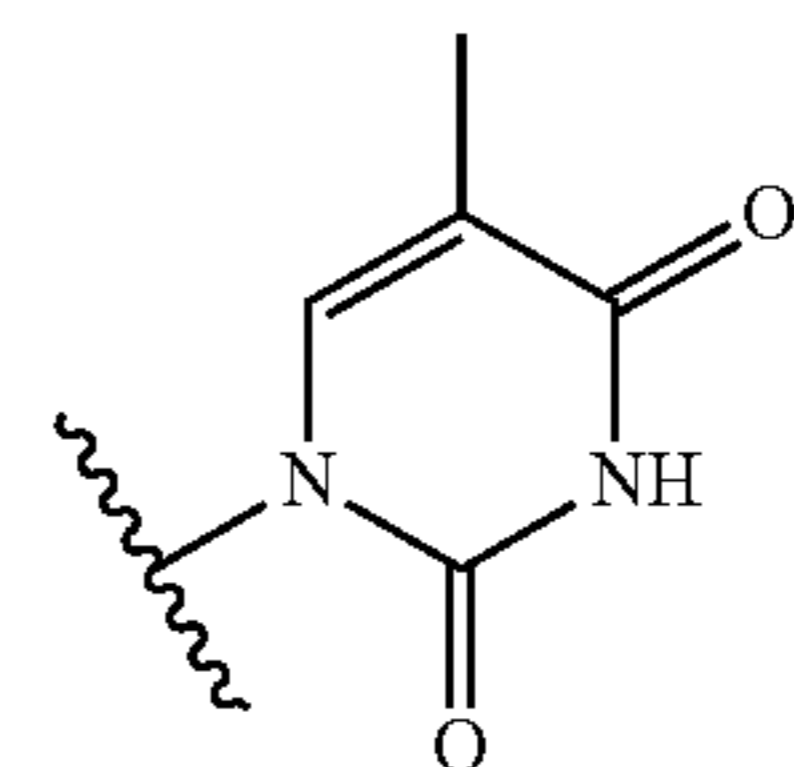
R⁴ is, independently for each occurrence, selected from the group consisting of:



(PC)



(DPG)



(T)

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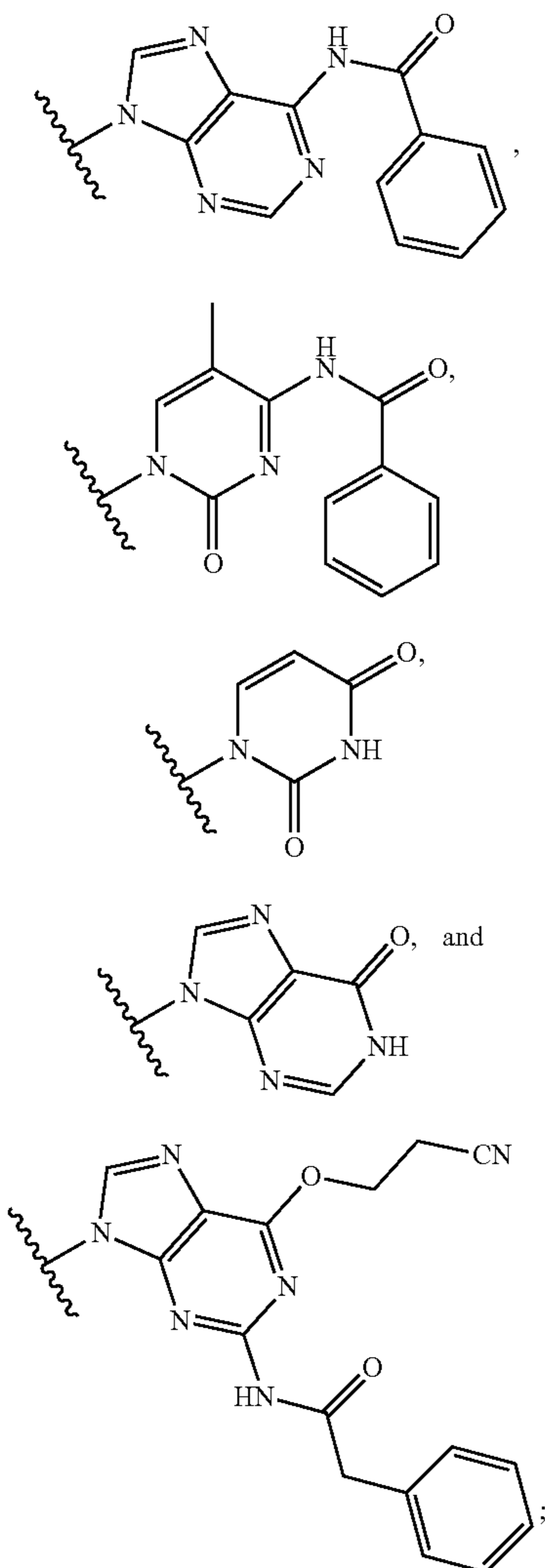
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243

-continued



and

(j) contacting the compound of Formula (B11) with a deprotecting agent to form the oligomeric compound of Formula (B).

In some embodiments, step (d) or step (e2) further comprises contacting the compound of Formula (IV-B) or the compound formed by the immediately prior step, respectively, with a capping agent.

In some embodiments, each step is performed in the presence of at least one solvent.

In some embodiments, the deblocking agent used in each step is a solution comprising a halogenated acid. In some embodiments, the deblocking agent used in each step is cyanoacetic acid. In some embodiments, the halogenated acid is selected from the group consisting of chloroacetic acid, dichloroacetic acid, trichloroacetic acid, fluoroacetic acid, difluoroacetic acid, and trifluoroacetic acid. In some embodiments, the halogenated acid is trifluoroacetic acid.

In some embodiments, at least one of steps (a), (c), (e1), and (f) further comprise the step of contacting the deblocked compound of each step with a neutralization agent. In some embodiments, each of steps (a), (c), (e1), and (f) further comprise the step of contacting the deblocked compound of each step with a neutralization agent. In some embodiments, the neutralization agent is in a solution comprising dichlo-

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romethane and isopropyl alcohol. In some embodiments, the neutralization agent is a monoalkyl, dialkyl, or trialkyl amine. In some embodiments, the neutralization agent is N,N-diisopropylethylamine.

In some embodiments, the deblocking agent used in each step is a solution comprising 4-cyanopyridine, dichloromethane, trifluoroacetic acid, trifluoroethanol, and water.

In some embodiments, the capping agent is in a solution comprising ethylmorpholine and methylpyrrolidinone. In some embodiments, the capping agent is an acid anhydride. In some embodiments, the acid anhydride is benzoic anhydride.

In some embodiments, the compounds of Formula (B4) and Formula (B8) are each, independently, in a solution comprising ethylmorpholine and dimethylimidazolidinone.

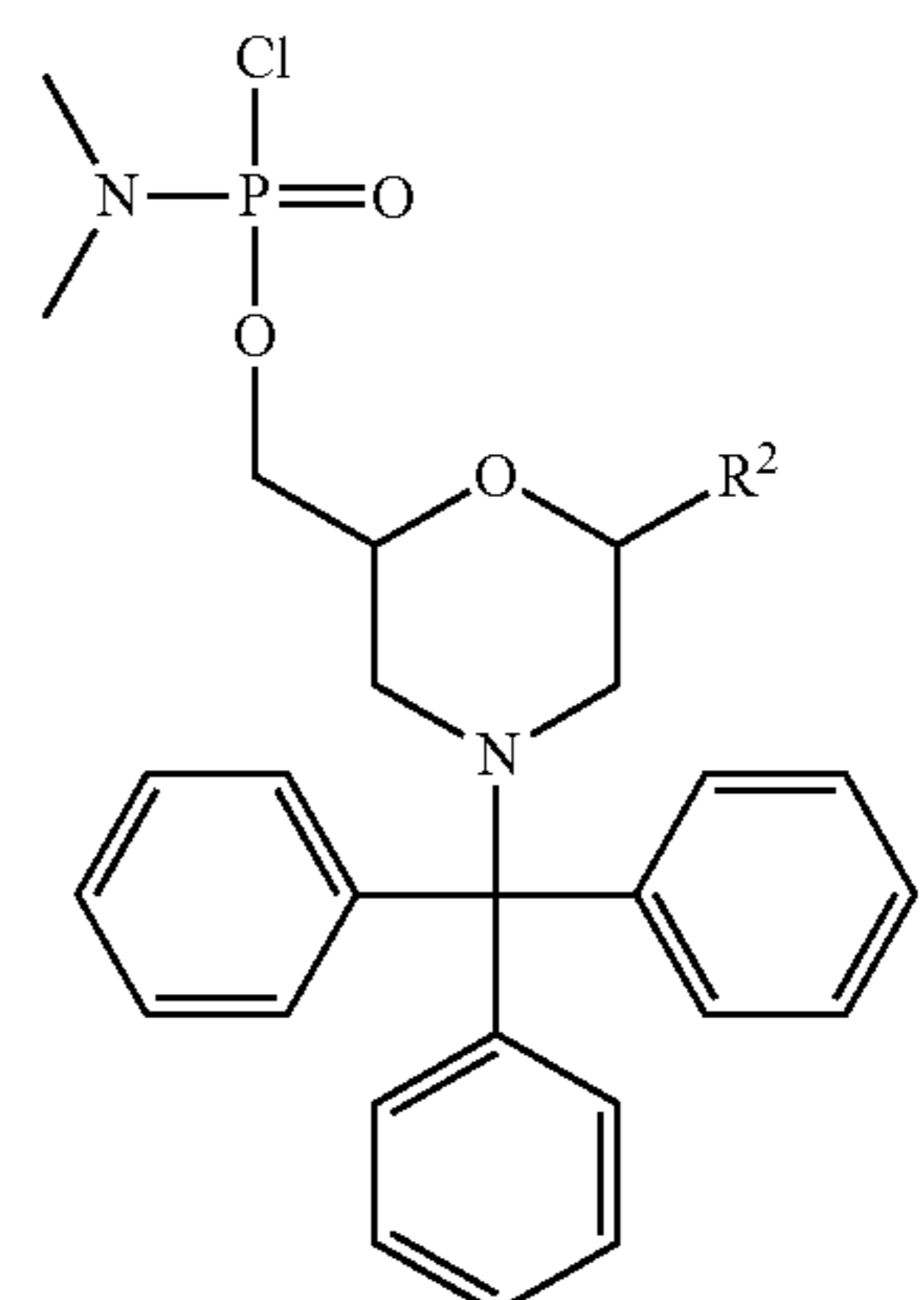
In some embodiments, the cleavage agent comprises dithiothreitol and 1,8-diazabicyclo[5.4.0]undec-7-ene. In some embodiments, the cleavage agent is in a solution comprising N-methyl-2-pyrrolidone.

In some embodiments, the deprotecting agent comprises NH_3 . In some embodiments, the deprotecting agent is in an aqueous solution.

In some embodiments, the support-medium comprises polystyrene with 1% crosslinked divinylbenzene.

Oligomeric compounds of the disclosure may have asymmetric centers, chiral axes, and chiral planes (as described, for example, in: E. L. Eliel and S. H. Wilen, *Stereochemistry of Carbon Compounds*, John Wiley & Sons, New York, 1994, pages 1119-1190, and March, J., *Advanced Organic Chemistry*, 3d. Ed., Chap. 4, John Wiley & Sons, New York (1985)), and may occur as racemates, racemic mixtures, and as individual diastereomers, with all possible isomers and mixtures thereof, including optical isomers. Oligomeric compounds of the disclosure herein specifically mentioned, without any indication of its stereo-chemistry, are intended to represent all possible isomers and mixtures thereof.

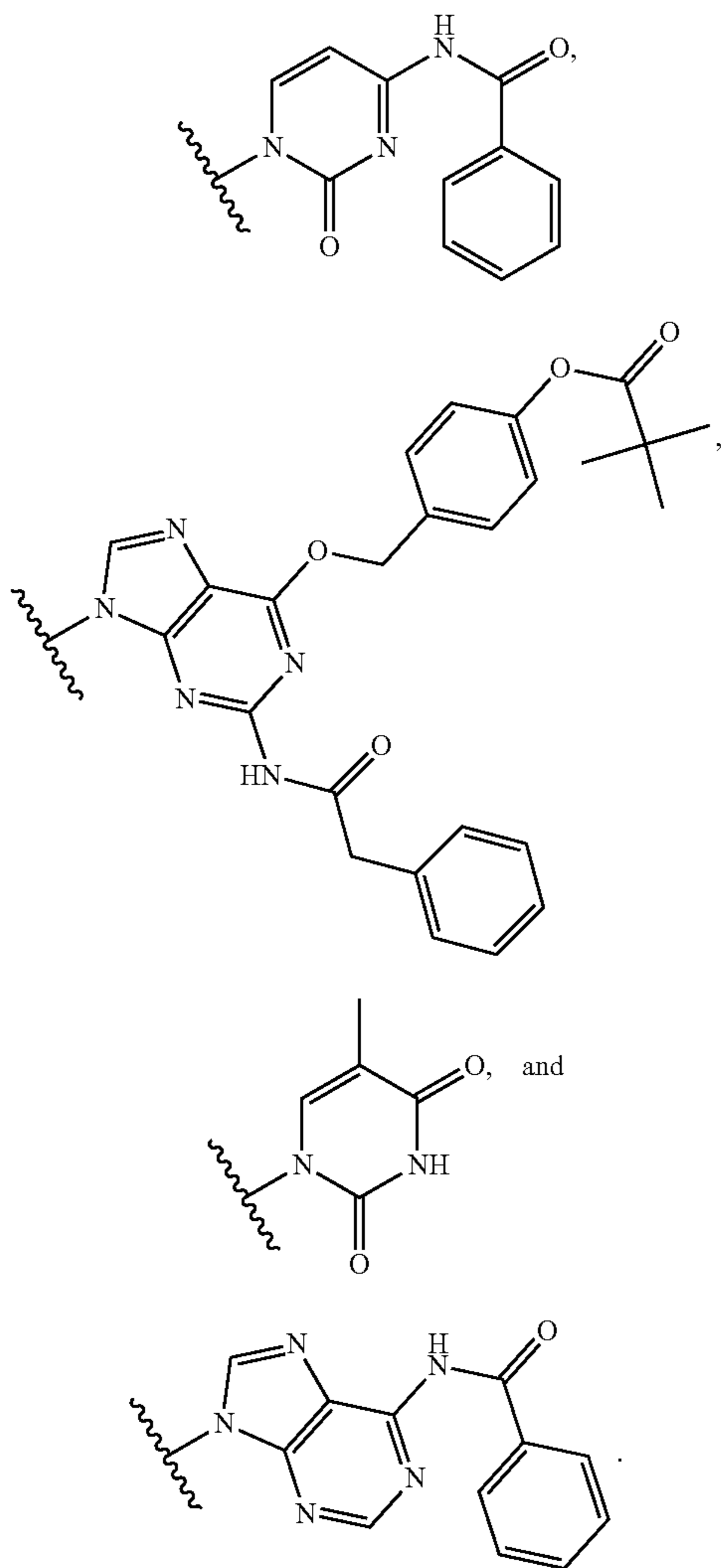
Specifically, without wishing to be bound by any particular theory, oligomeric compounds of the disclosure are prepared, as discussed herein, from activated morpholino subunits including such non-limiting examples such as a compound of Formula (VIII):



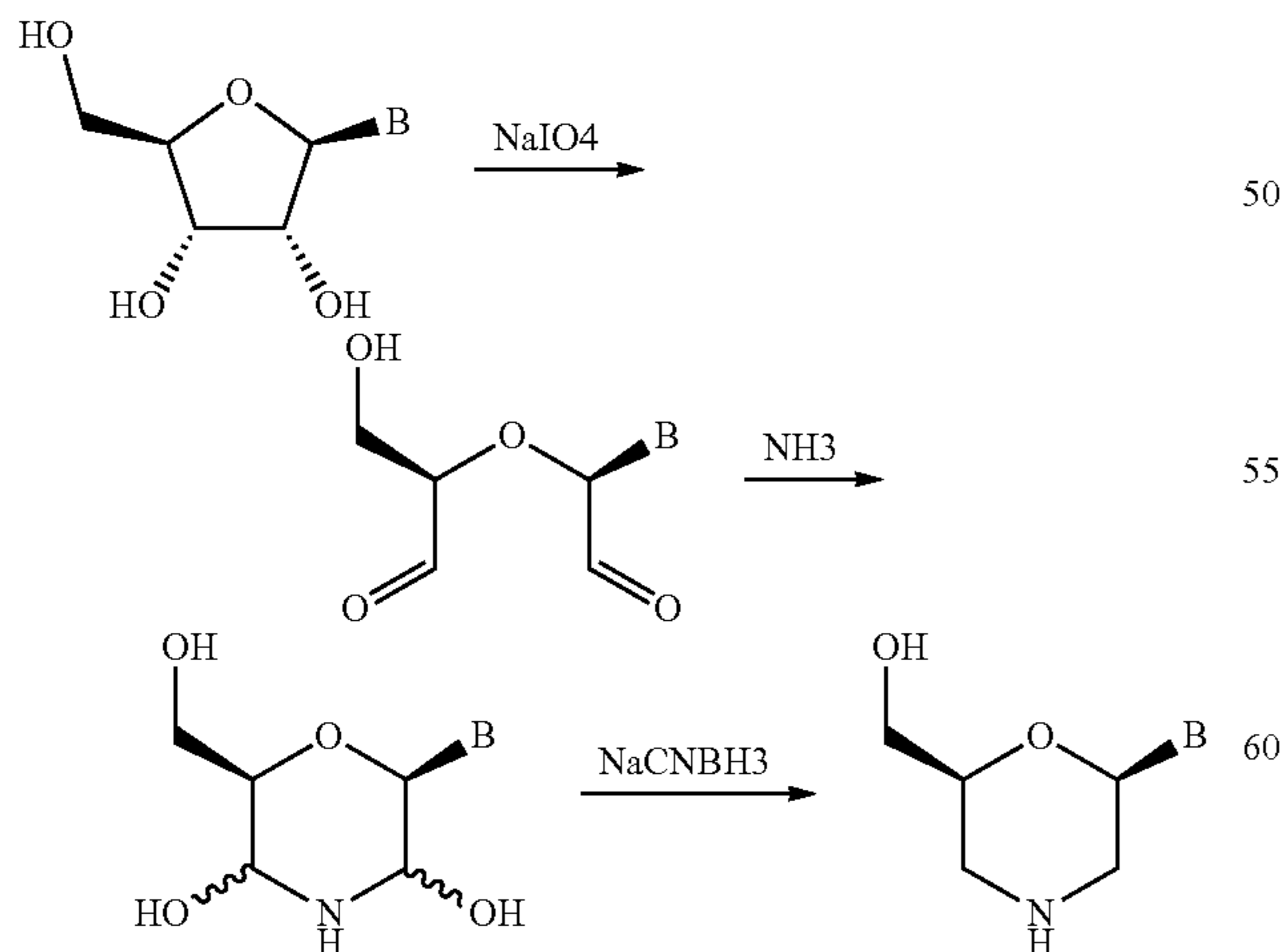
(VIII)

wherein R^2 is, independently for each compound of Formula (VIII), selected from the group consisting of:

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Each of the above-mentioned compounds of Formula (VIII) may be prepared, for example, from the corresponding beta-D-ribofuranosyl as depicted below:

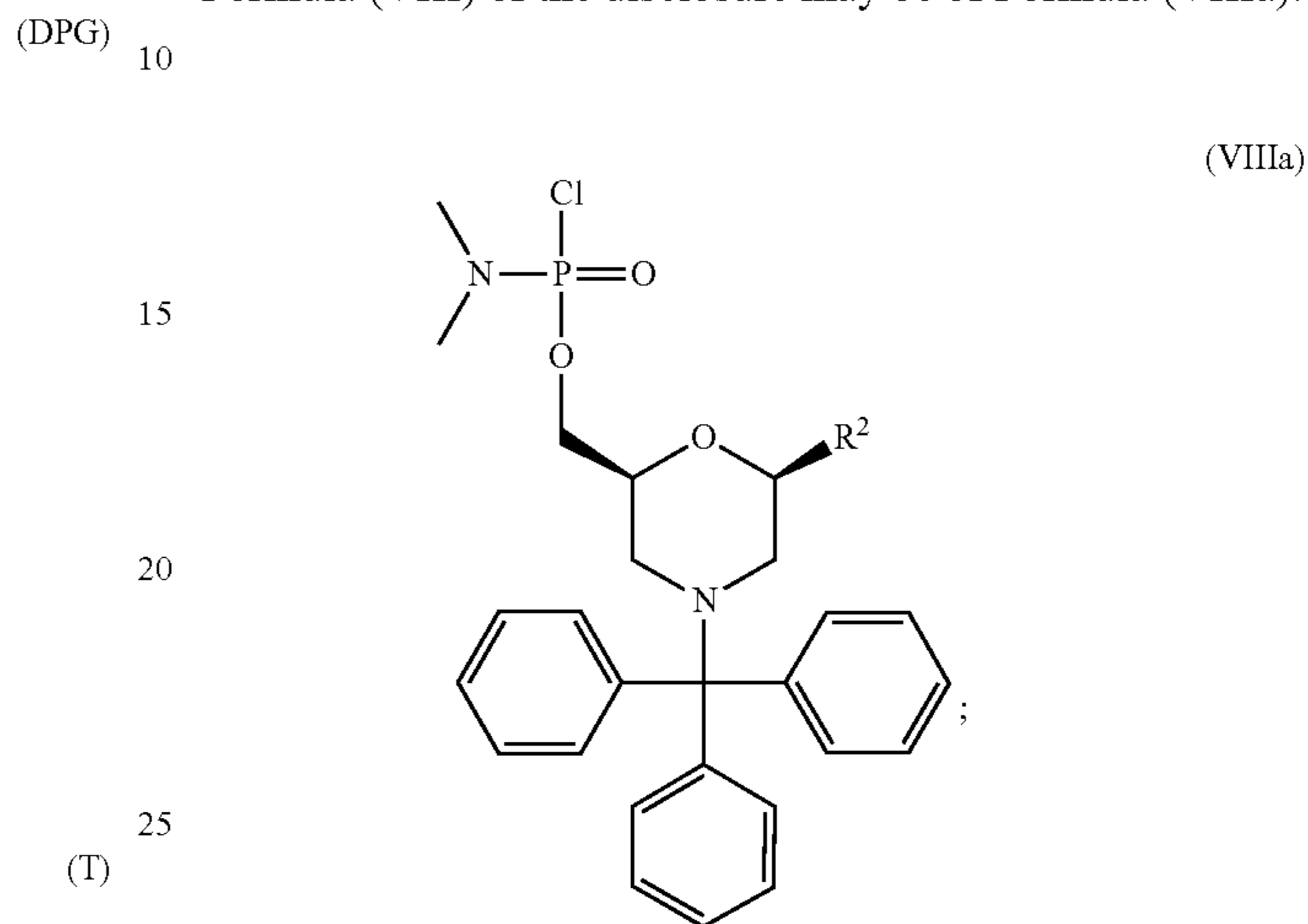


See Summerton et al., *Antisense Nucleic Acid Drug Dev.* 1997, 7, 187-195. Without being bound by any particular

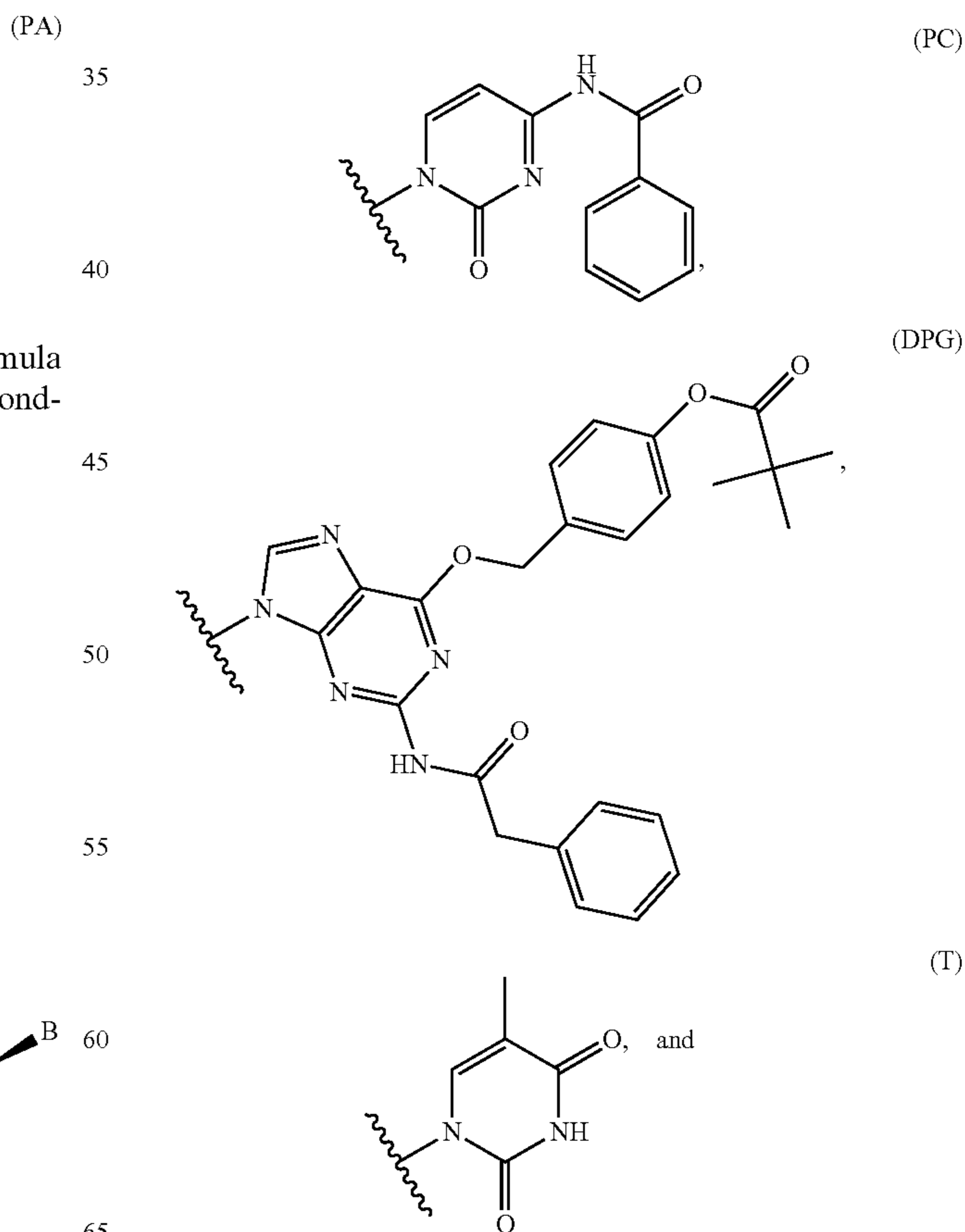
246

(PC) theory, the stereochemistry of the two chiral carbons is retained under the synthetic conditions such that a number of possible stereoisomers of each morpholino subunit may be produced based on selection of, for example, an alpha-L-ribofuranosyl, alpha-D-ribofuranosyl, beta-L-ribofuranosyl, or beta-D-ribofuranosyl starting material.

For example, in some embodiments, a compound of Formula (VIII) of the disclosure may be of Formula (VIIIa):



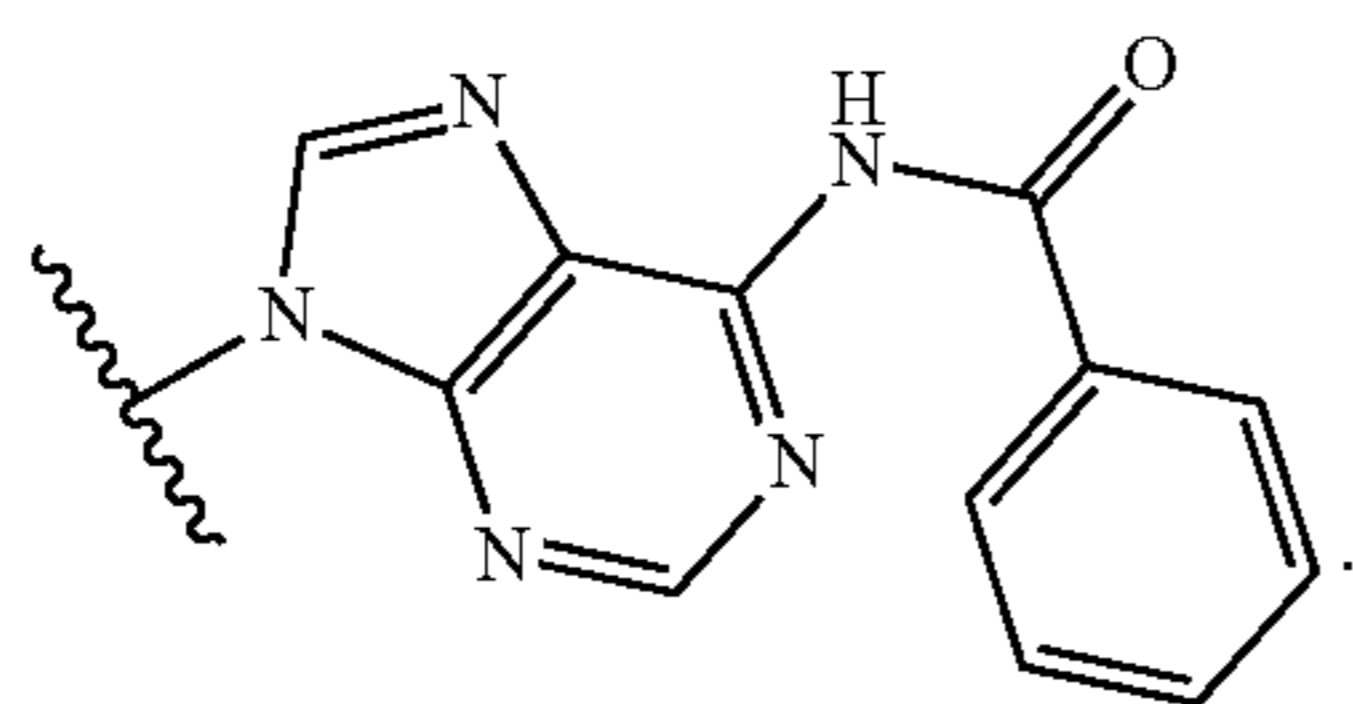
wherein R² is, independently for each compound of Formula (VIIIa), selected from the group consisting of:



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-continued



Without being bound by any particular theory, incorporation of 10 to 40 compounds of Formula (VIII), for example, into an oligomeric compound of the disclosure may result in numerous possible stereoisomers.

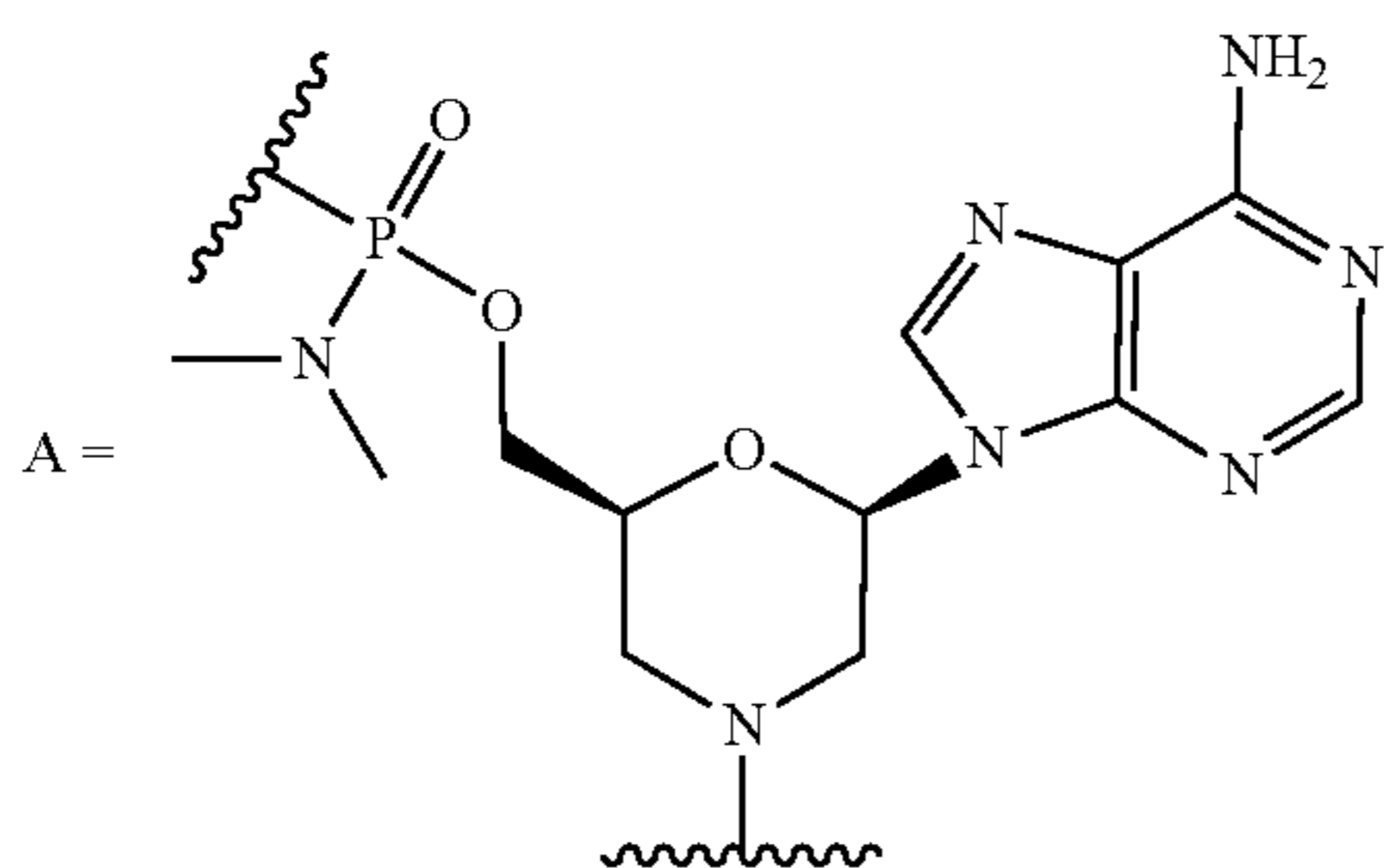
Without wishing to be bound by any particular theory, oligomeric compounds of the disclosure comprise one or more phosphorous-containing intersubunit linkages, which create a chiral center at each phosphorus, each of which is designated as either an “Sp” or “Rp” configuration as understood in the art. Without wishing to be bound by any particular theory, this chirality creates stereoisomers, which have identical chemical composition but different three-dimensional arrangement of their atoms.

Without wishing to be bound by any particular theory, the configuration of each phosphorous-containing intersubunit linkage occurs randomly during synthesis of, for example, oligomeric compounds of the disclosure. Without wishing to be bound by any particular theory, the synthesis process generates an exponentially large number of stereoisomers of an oligomeric compound of the disclosure because oligomeric compounds of the disclosure are comprised of numerous phosphorous-containing intersubunit linkages—with each phosphorous-containing intersubunit linkage having a random chiral configuration. Specifically, without wishing to be bound by any particular theory, each intersubunit linkage of an additional morpholino subunit doubles the number of stereoisomers of the product, so that a conventional preparation of an oligomeric compound of the disclosure is in fact a highly heterogeneous mixtures of 2^N stereoisomers, where N represents the number of phosphorous-containing intersubunit linkages.

Table 1 depicts various embodiments of morpholino subunits provided in the processes described herein.

TABLE 1

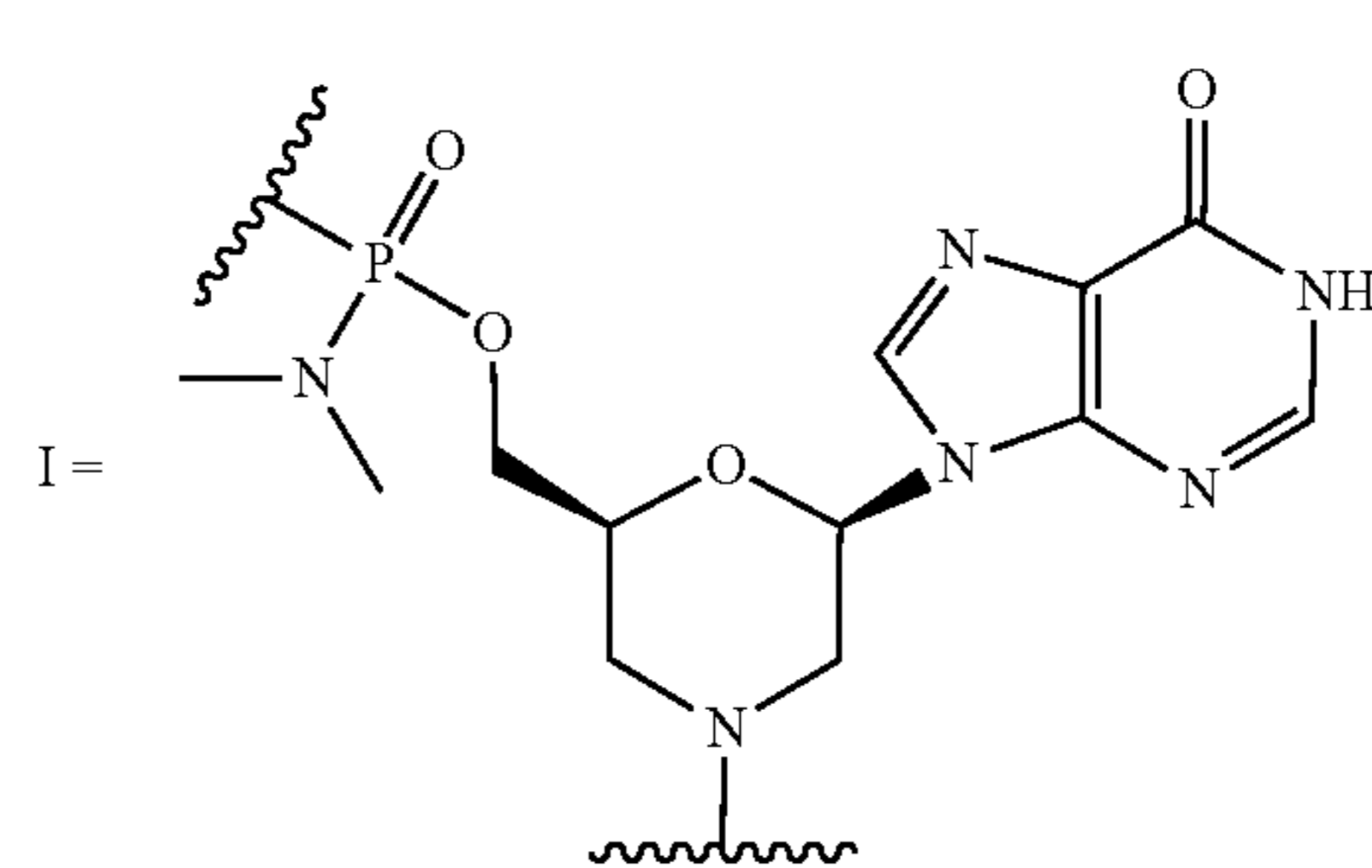
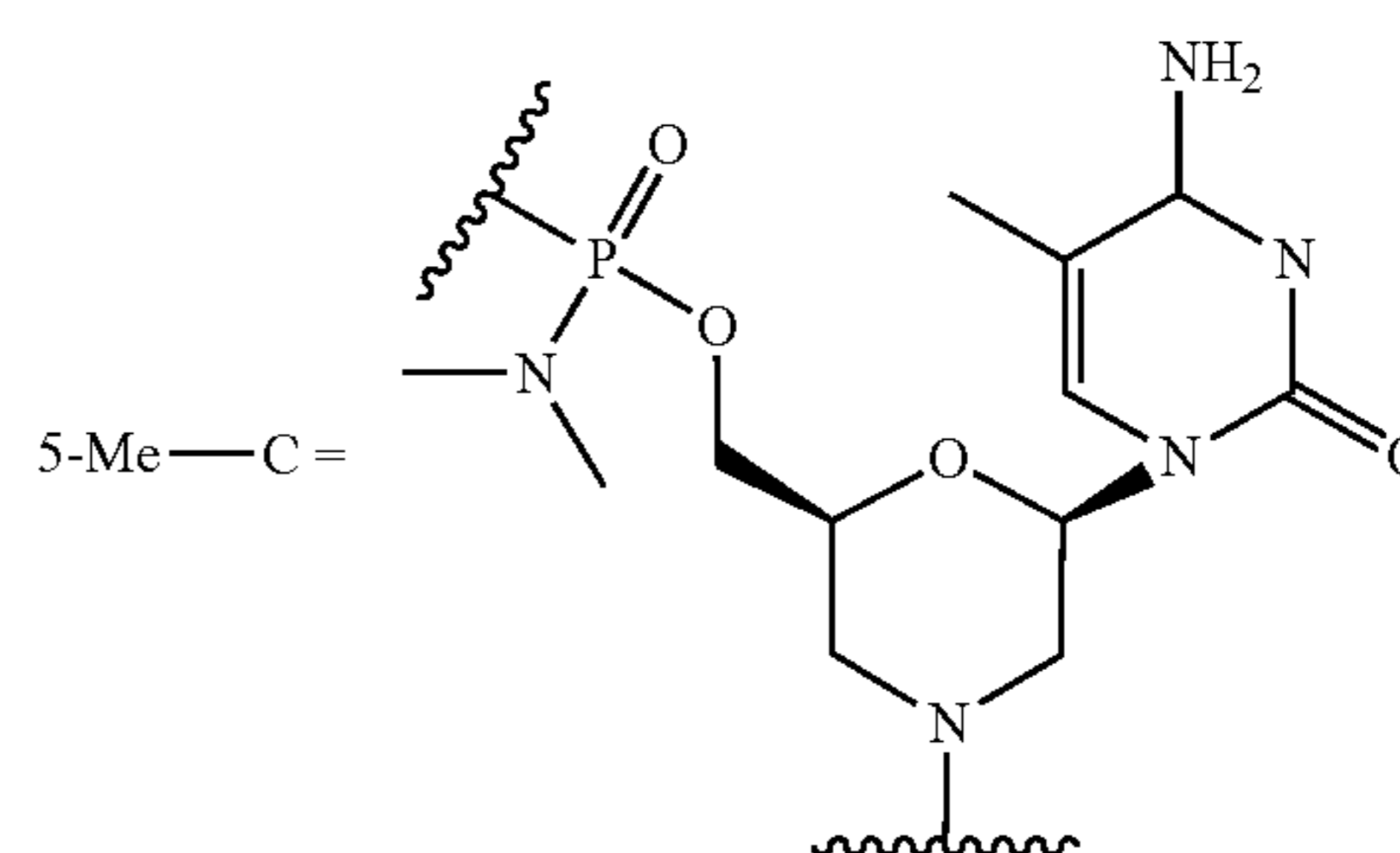
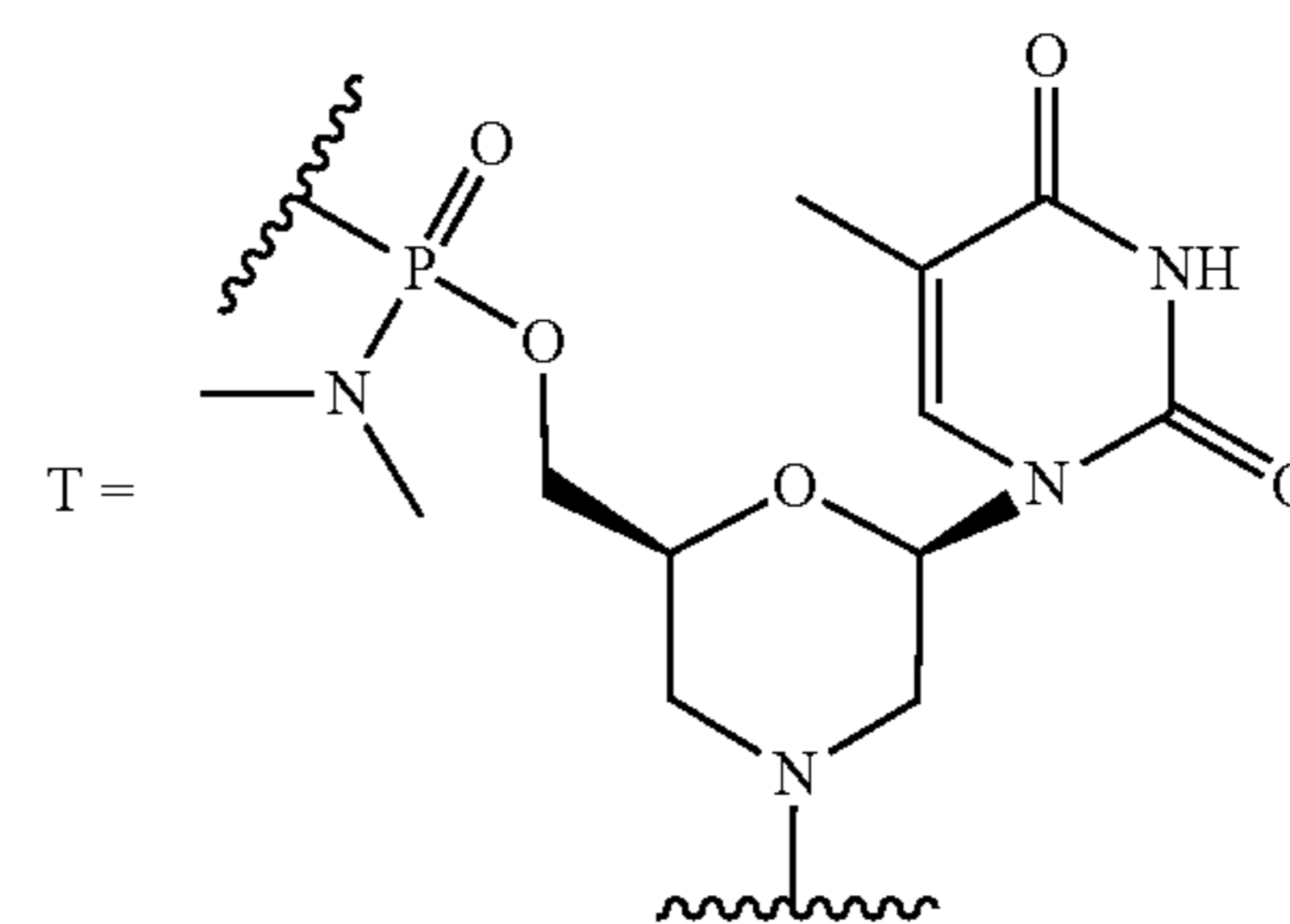
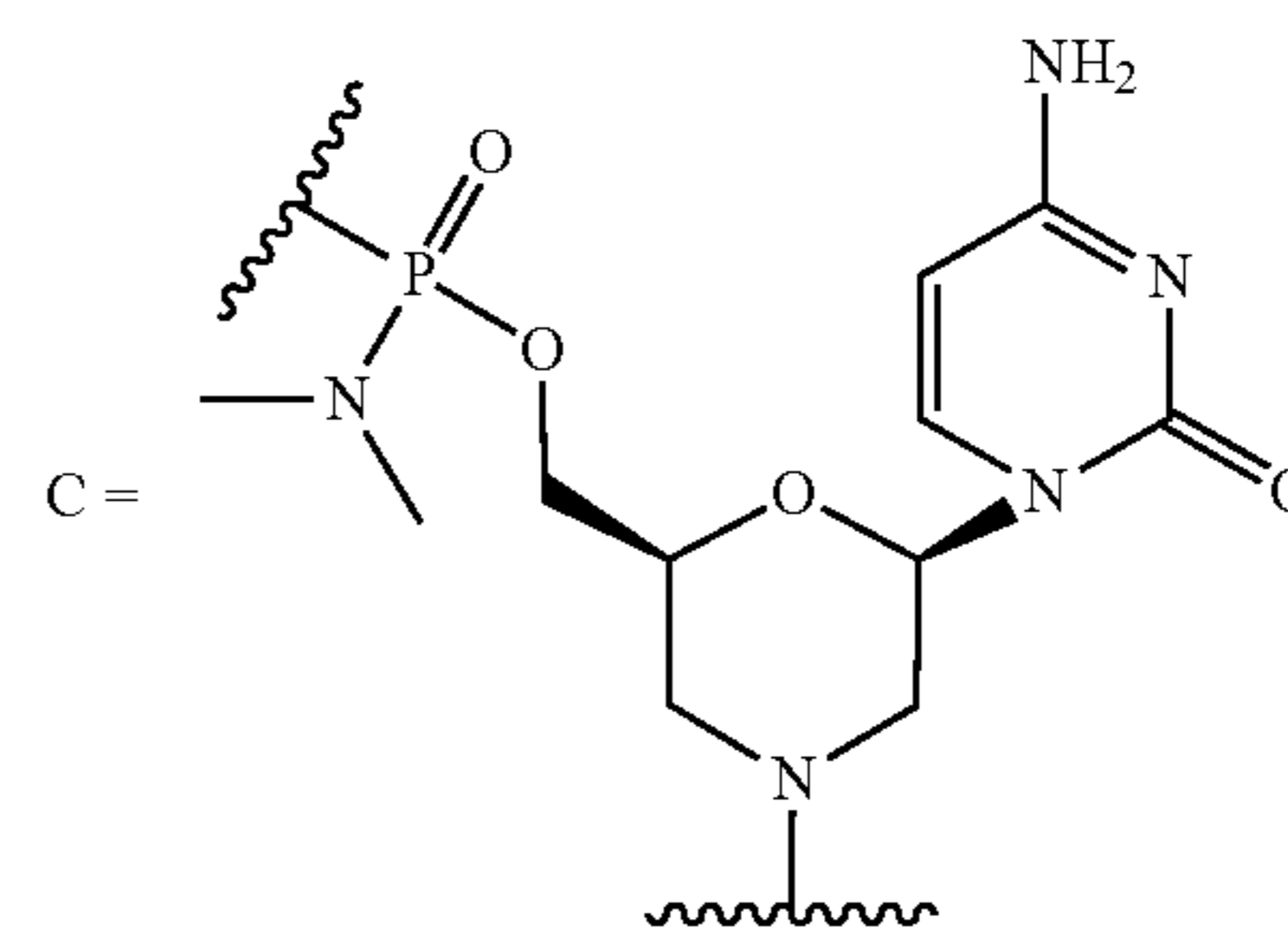
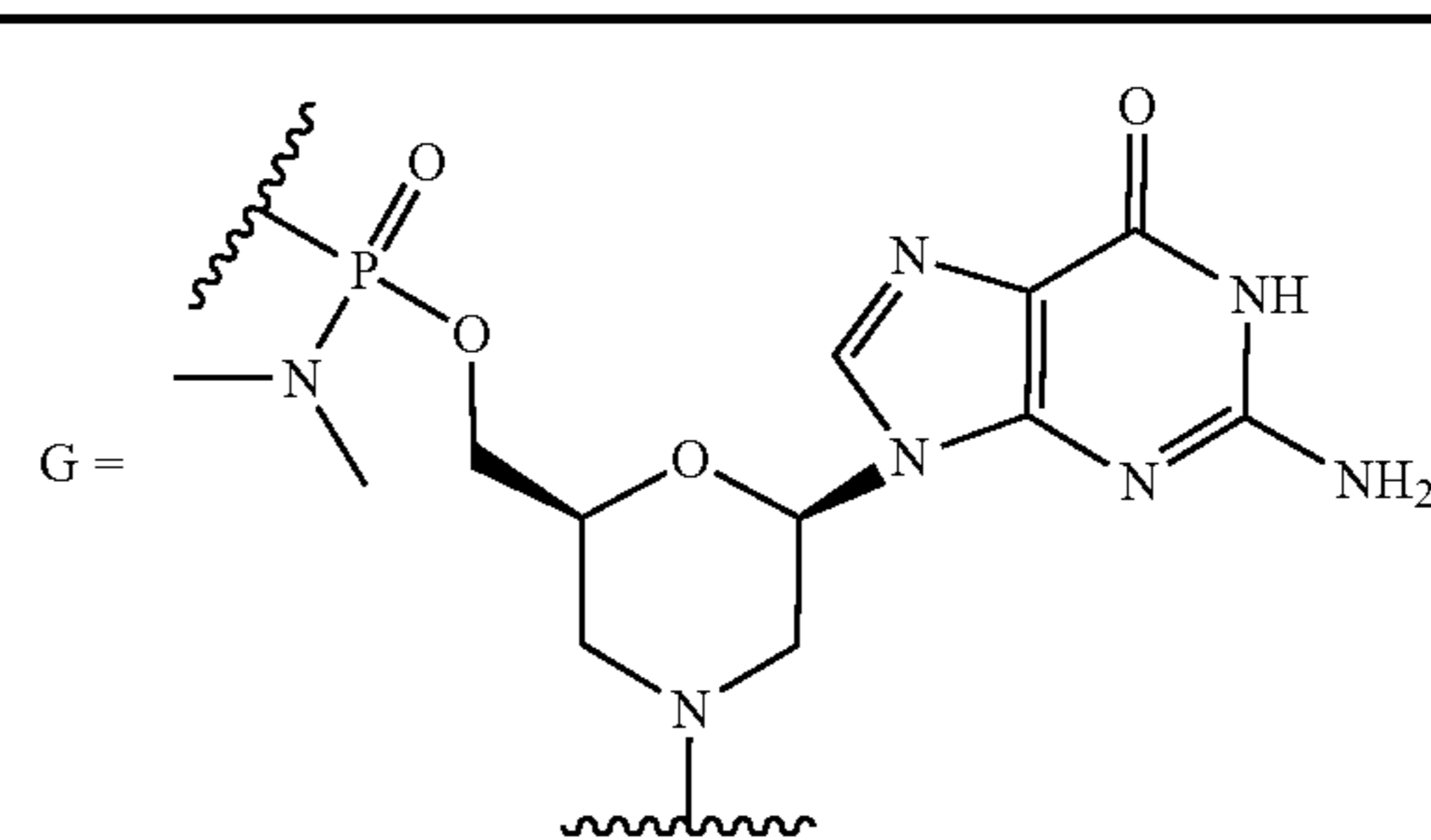
Various embodiments of morpholino subunits.



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TABLE 1-continued

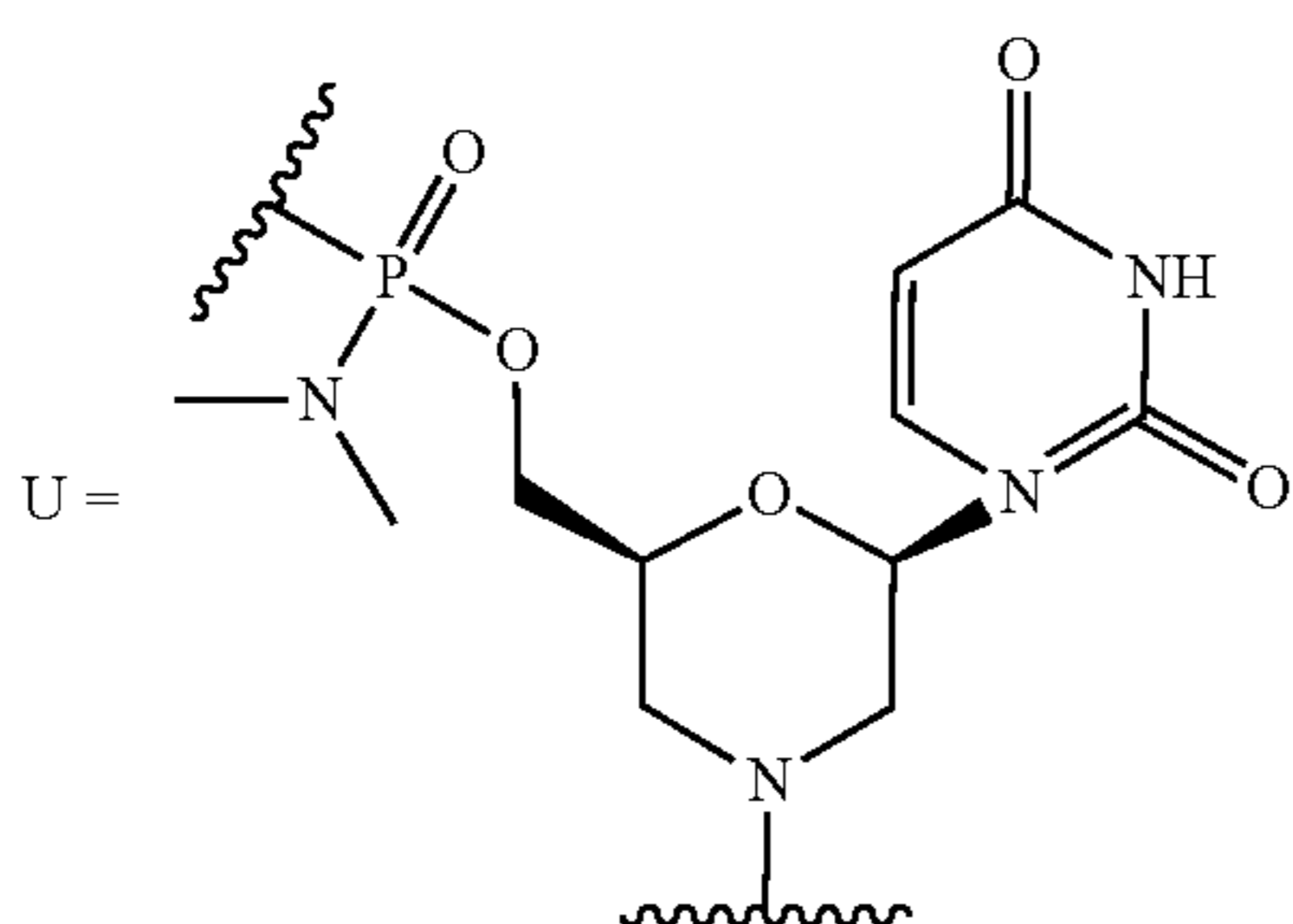
Various embodiments of morpholino subunits.



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TABLE 1-continued

Various embodiments of morpholino subunits.

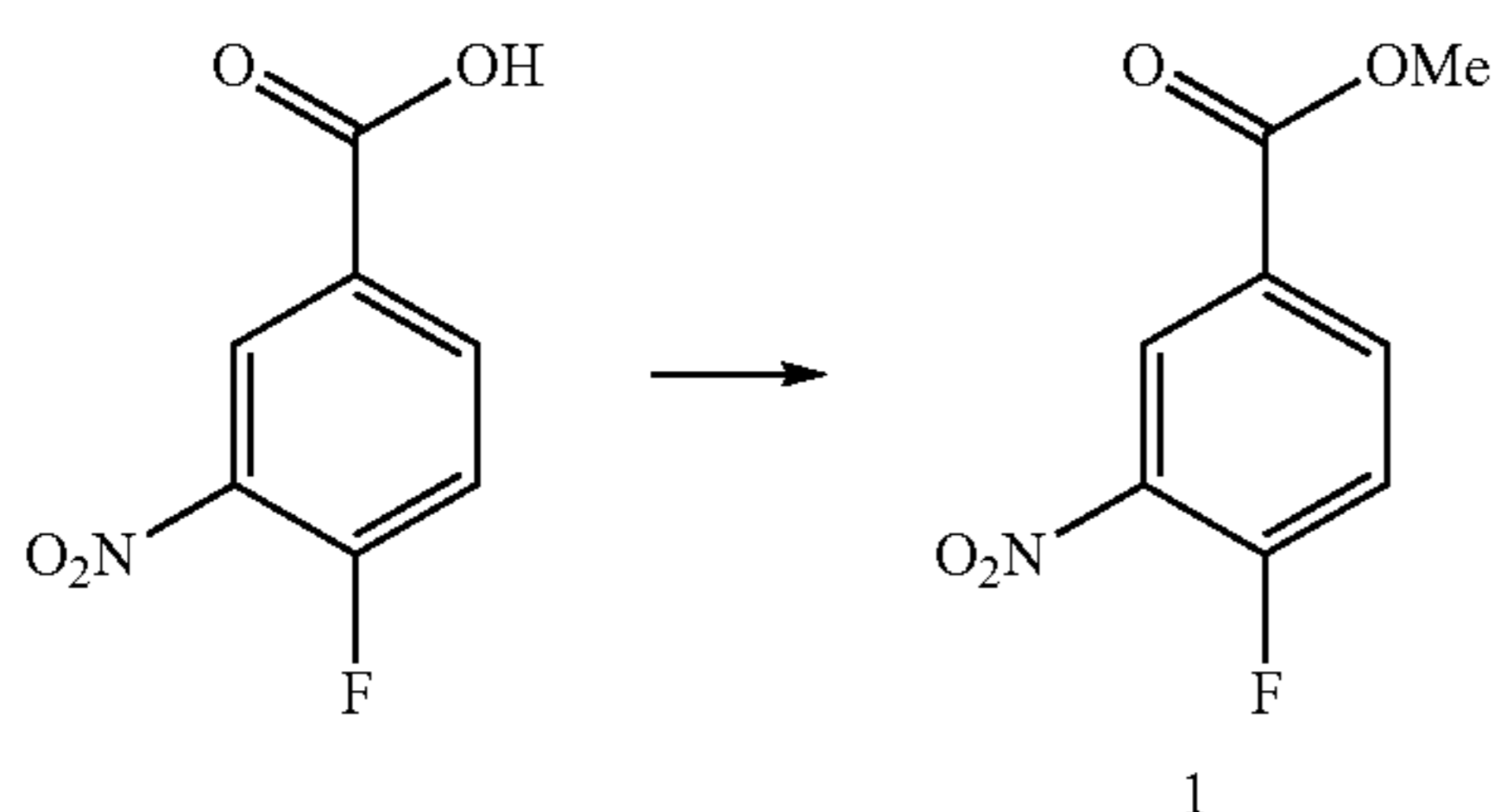


EXAMPLES

Examples have been set forth below for the purpose of illustration and to describe certain specific embodiments of the disclosure. However, the scope of the claims is not to be in any way limited by the examples set forth herein. Various changes and modifications to the disclosed embodiments will be apparent to those skilled in the art and such changes and modifications including, without limitation, those relating to the chemical structures, substituents, derivatives, formulations or methods of the disclosure may be made without departing from the spirit of the disclosure and the scope of the appended claims. Definitions of the variables in the structures in the schemes herein are commensurate with those of corresponding positions in the formulae presented herein.

Example 1: NCP2 Anchor Synthesis

1. Preparation of Methyl 4-Fluoro-3-Nitrobenzoate (1)

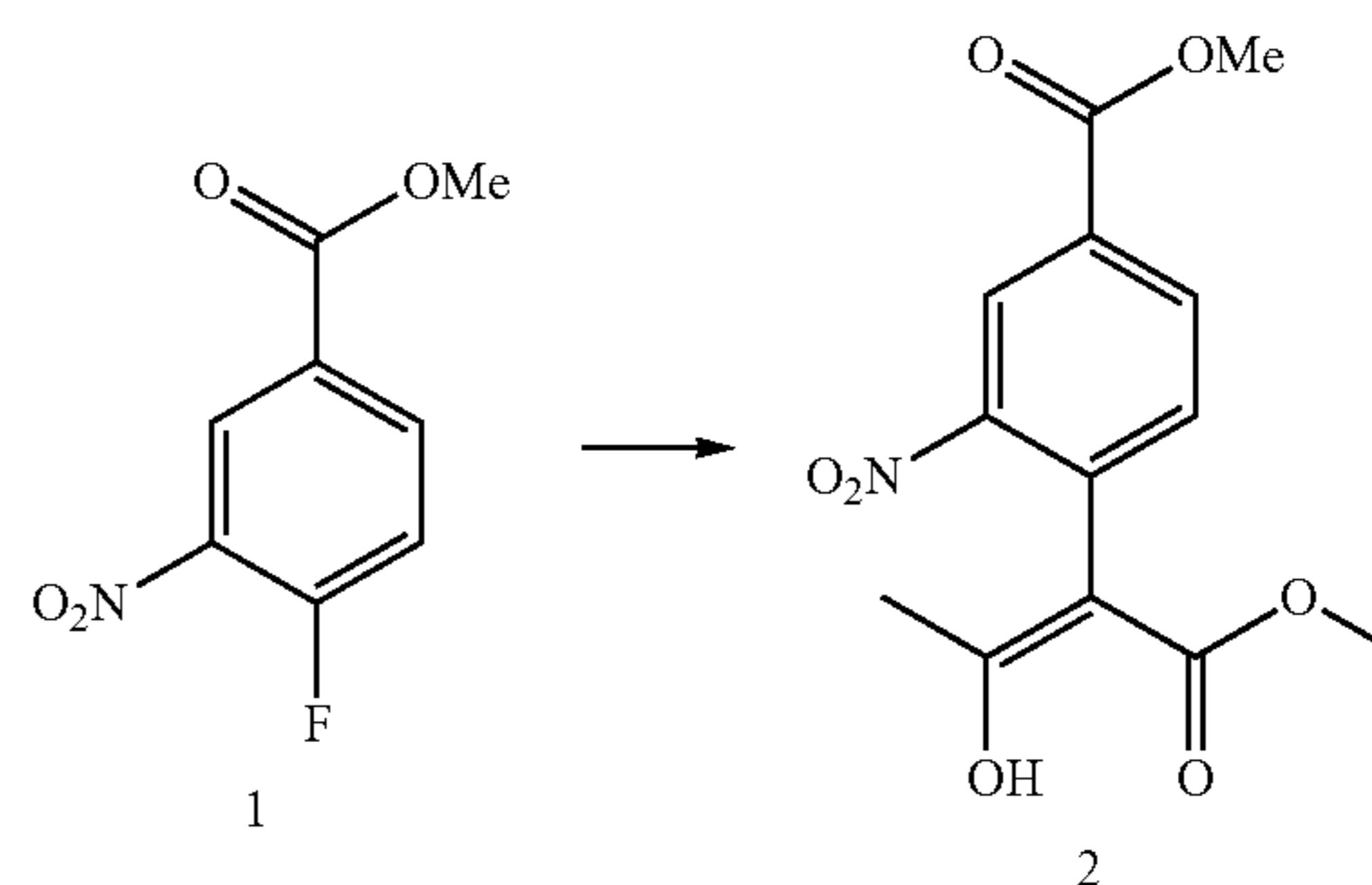


To a 100 L flask was charged 12.7 kg of 4-fluoro-3-nitrobenzoic acid was added 40 kg of methanol and 2.82 kg concentrated sulfuric acid. The mixture was stirred at reflux (65° C.) for 36 hours. The reaction mixture was cooled to 0° C. Crystals formed at 38° C. The mixture was held at 0° C. for 4 hrs then filtered under nitrogen. The 100 L flask was washed and filter cake was washed with 10 kg of methanol that had been cooled to 0° C. The solid filter cake was dried on the funnel for 1 hour, transferred to trays, and dried in a vacuum oven at room temperature to a constant weight of 13.695 kg methyl 4-fluoro-3-nitrobenzoate (100% yield; HPLC 99%).

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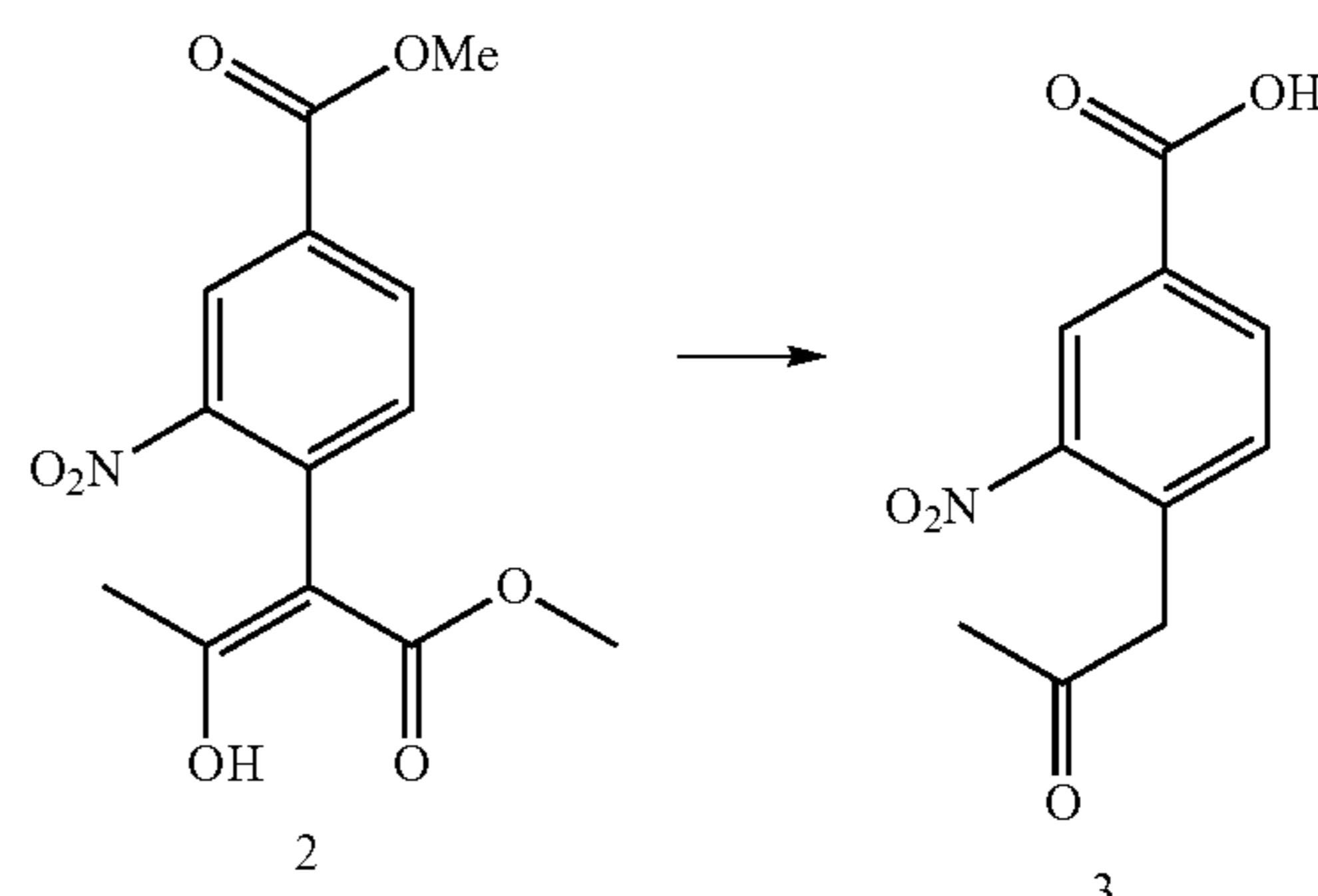
2. Preparation of 3-Nitro-4-(2-oxopropyl)benzoic Acid

A. (Z)-Methyl 4-(3-Hydroxy-1-Methoxy-1-Oxobut-2-en-2-yl)-3-Nitrobenzoate (2)



To a 100 L flask was charged 3.98 kg of methyl 4-fluoro-3-nitrobenzoate (1) from the previous step 9.8 kg DMF, 2.81 kg methyl acetoacetate. The mixture was stirred and cooled to 0° C. To this was added 3.66 kg DBU over about 4 hours while the temperature was maintained at or below 5° C. The mixture was stirred an additional 1 hour. To the reaction flask was added a solution of 8.15 kg of citric acid in 37.5 kg of purified water while the reaction temperature was maintained at or below 15° C. After the addition, the reaction mixture was stirred an addition 30 minutes then filtered under nitrogen. The wet filter cake was returned to the 100 L flask along with 14.8 kg of purified water. The slurry was stirred for 10 minutes then filtered. The wet cake was again returned to the 100 L flask, slurried with 14.8 kg of purified water for 10 minutes, and filtered to crude (Z)-methyl 4-(3-hydroxy-1-methoxy-1-oxobut-2-en-2-yl)-3-nitrobenzoate.

B. 3-Nitro-4-(2-oxopropyl)benzoic Acid

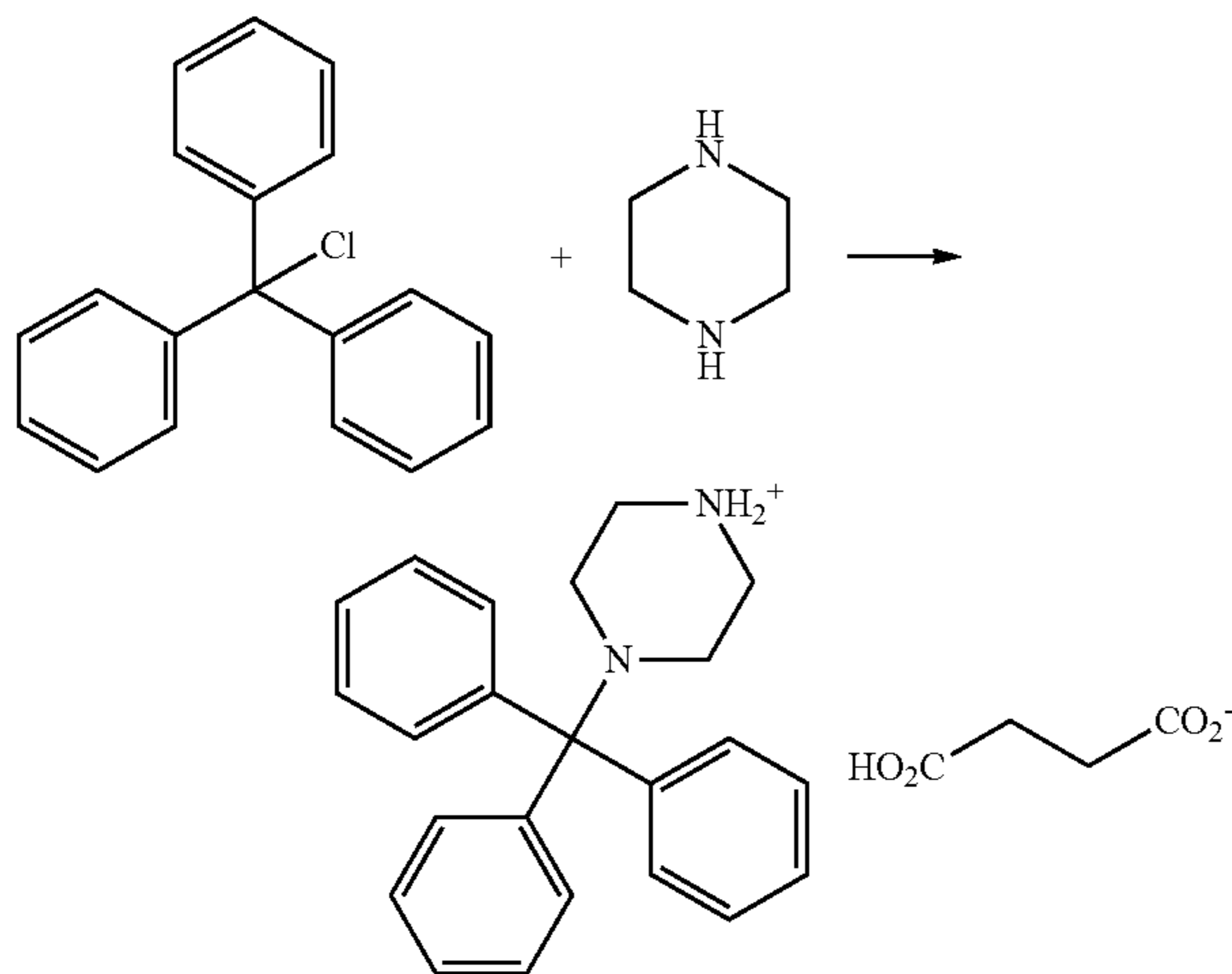


The crude (Z)-methyl 4-(3-hydroxy-1-methoxy-1-oxobut-2-en-2-yl)-3-nitrobenzoate was charged to a 100 L reaction flask under nitrogen. To this was added 14.2 kg 1,4-dioxane and the stirred. To the mixture was added a solution of 16.655 kg concentrated HCl and 13.33 kg purified water (6M HCl) over 2 hours while the temperature of the reaction mixture was maintained below 15° C. When the addition was complete, the reaction mixture was heated at reflux (80° C.) for 24 hours, cooled to room temperature, and filtered under nitrogen. The solid filter cake was tritu-

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rated with 14.8 kg of purified water, filtered, triturated again with 14.8 kg of purified water, and filtered. The solid was returned to the 100 L flask with 39.9 kg of DCM and refluxed with stirring for 1 hour. 1.5 kg of purified water was added to dissolve the remaining solids. The bottom organic layer was split to a pre-warmed 72 L flask, then returned to a clean dry 100 L flask. The solution was cooled to 0° C., held for 1 hour, then filtered. The solid filter cake was washed twice each with a solution of 9.8 kg DCM and 5 kg heptane, then dried on the funnel. The solid was transferred to trays and dried to a constant weight of 1.855 kg 3-Nitro-4-(2-oxopropyl)benzoic Acid. Overall yield 42% from compound 1. HPLC 99.45%.

3. Preparation of N-Triethylpiperazine Succinate (NTP)



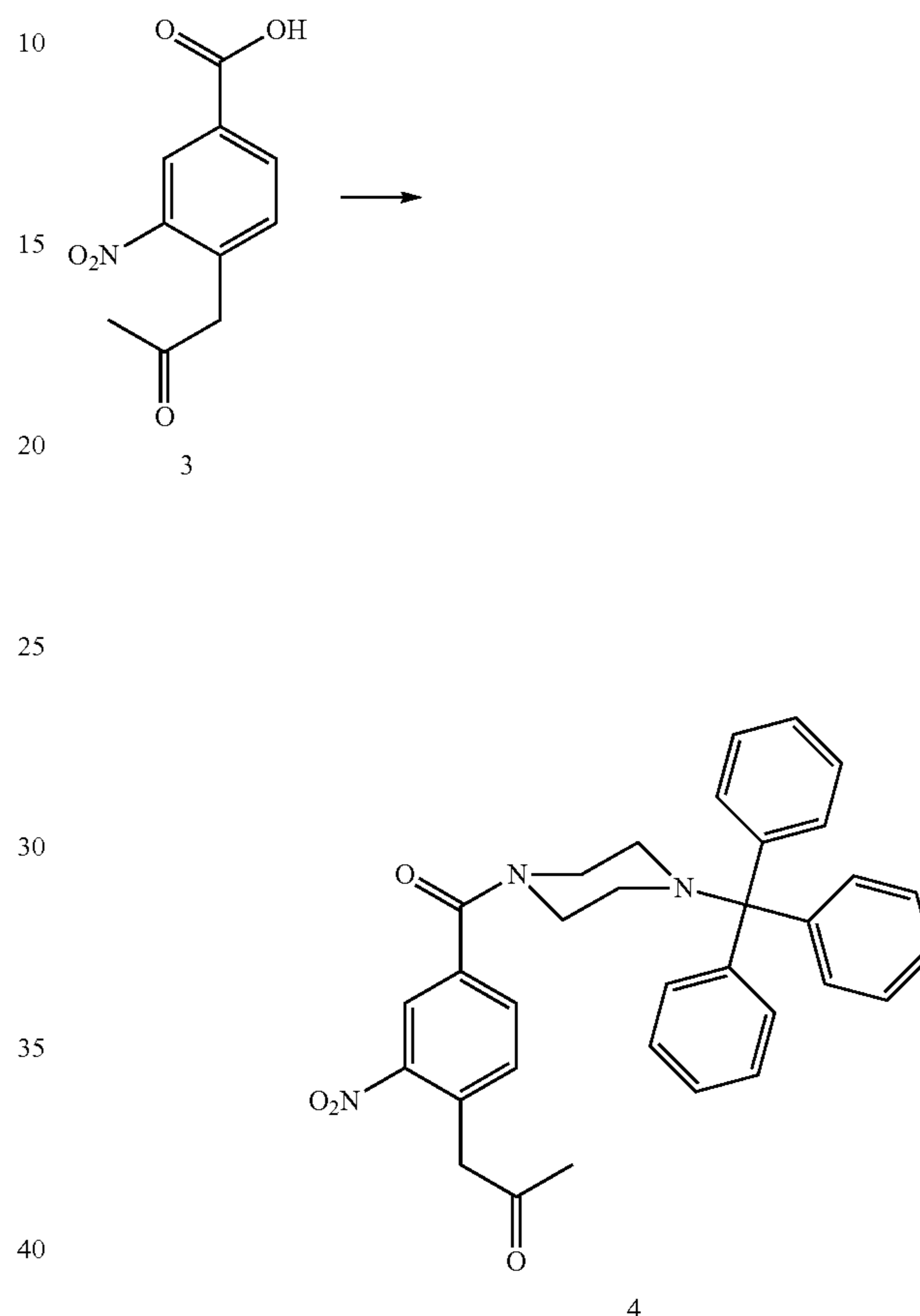
To a 72 L jacketed flask was charged under nitrogen 1.805 kg triphenylmethyl chloride and 8.3 kg of toluene (TPC solution). The mixture was stirred until the solids dissolved. To a 100 L jacketed reaction flask was added under nitrogen 5.61 kg piperazine, 19.9 kg toluene, and 3.72 kg methanol. The mixture was stirred and cooled to 0° C. To this was slowly added in portions the TPC solution over 4 hours while the reaction temperature was maintained at or below 10° C. The mixture was stirred for 1.5 hours at 10° C., then allowed to warm to 14° C. 32.6 kg of purified water was charged to the 72 L flask, then transferred to the 100 L flask while the internal batch temperature was maintained at 20+/-5° C. The layers were allowed to split and the bottom aqueous layer was separated and stored. The organic layer was extracted three times with 32 kg of purified water each, and the aqueous layers were separated and combined with the stored aqueous solution.

The remaining organic layer was cooled to 18° C. and a solution of 847 g of succinic acid in 10.87 kg of purified water was added slowly in portions to the organic layer. The mixture was stirred for 1.75 hours at 20+/-5° C. The mixture was filtered, and the solids were washed with 2 kg TBME and 2 kg of acetone then dried on the funnel. The filter cake was triturated twice with 5.7 kg each of acetone and filtered and washed with 1 kg of acetone between triturations. The solid was dried on the funnel, then transferred to trays and dried in a vacuum oven at room temperature to a constant weight of 2.32 kg of NTP. Yield 80%.

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4. Preparation of (4-(2-Hydroxypropyl)-3-Nitrophenyl)(4-Triethylpiperazin-1-yl)Methanone

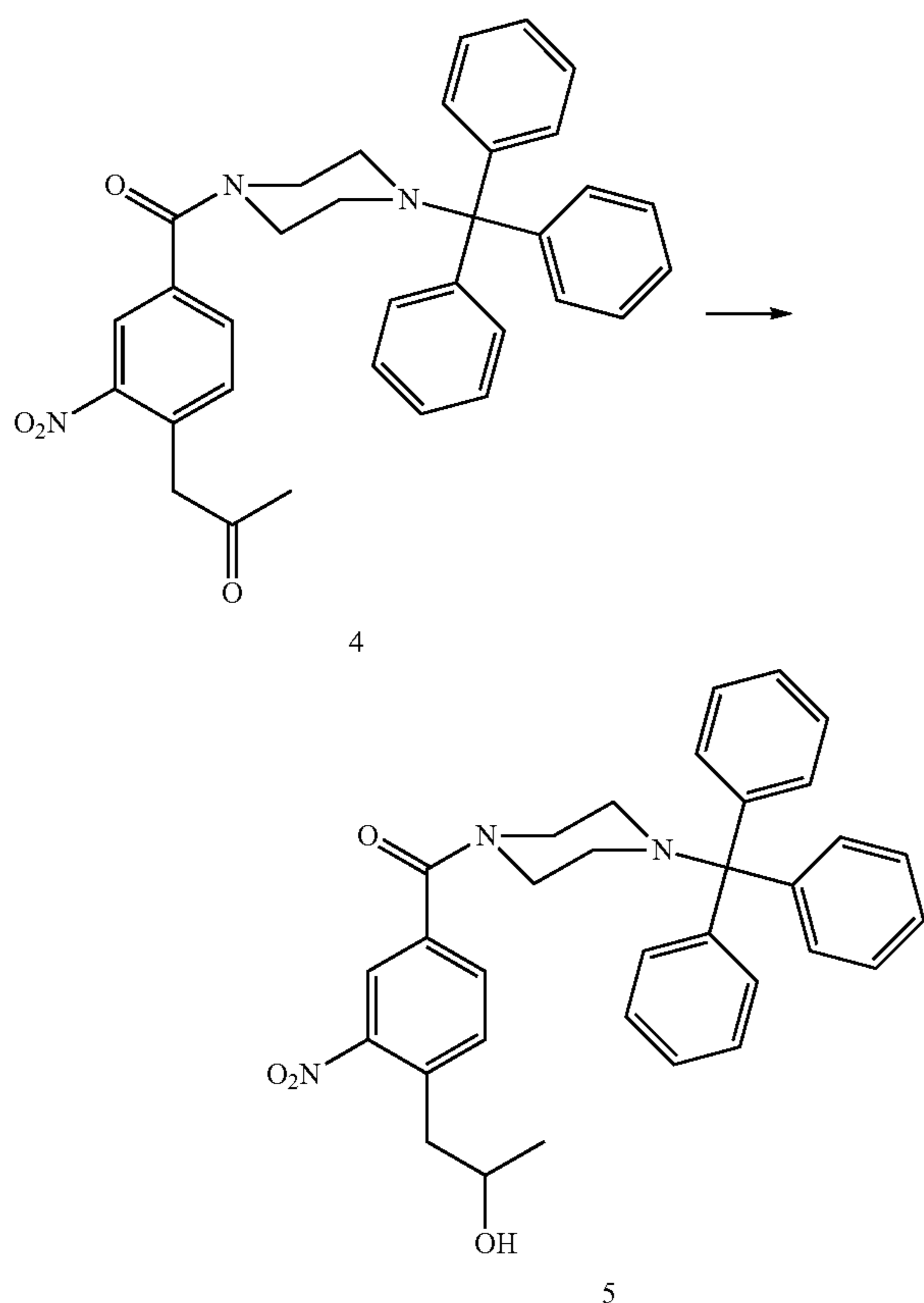
A. Preparation of 1-(2-Nitro-4(4-Triethylpiperazine-1-Carbonyl)Phenyl)Propan-2-one



To a 100 L jacketed flask was charged under nitrogen 2 kg of 3-Nitro-4-(2-oxopropyl)benzoic Acid (3), 18.3 kg DCM, 1.845 kg N-(3-dimethylaminopropyl)-N'-ethylcarbodiimide hydrochloride (EDC.HCl). The solution was stirred until a homogenous mixture was formed. 3.048 kg of NTP was added over 30 minutes at room temperature and stirred for 8 hours. 5.44 kg of purified water was added to the reaction mixture and stirred for 30 minutes. The layers were allowed to separate and the bottom organic layer containing the product was drained and stored. The aqueous layer was extracted twice with 5.65 kg of DCM. The combined organic layers were washed with a solution of 1.08 kg sodium chloride in 4.08 kg purified water. The organic layers were dried over 1.068 kg of sodium sulfate and filtered. The sodium sulfate was washed with 1.3 kg of DCM. The combined organic layers were slurried with 252 g of silica gel and filtered through a filter funnel containing a bed of 252 g of silica gel. The silica gel bed was washed with 2 kg of DCM. The combined organic layers were evaporated on a rotovap. 4.8 kg of THE was added to the residue and then evaporated on the rotovap until 2.5 volumes of the crude 1-(2-nitro-4(4-triethylpiperazine-1-carbonyl)phenyl)propan-2-one in THE was reached.

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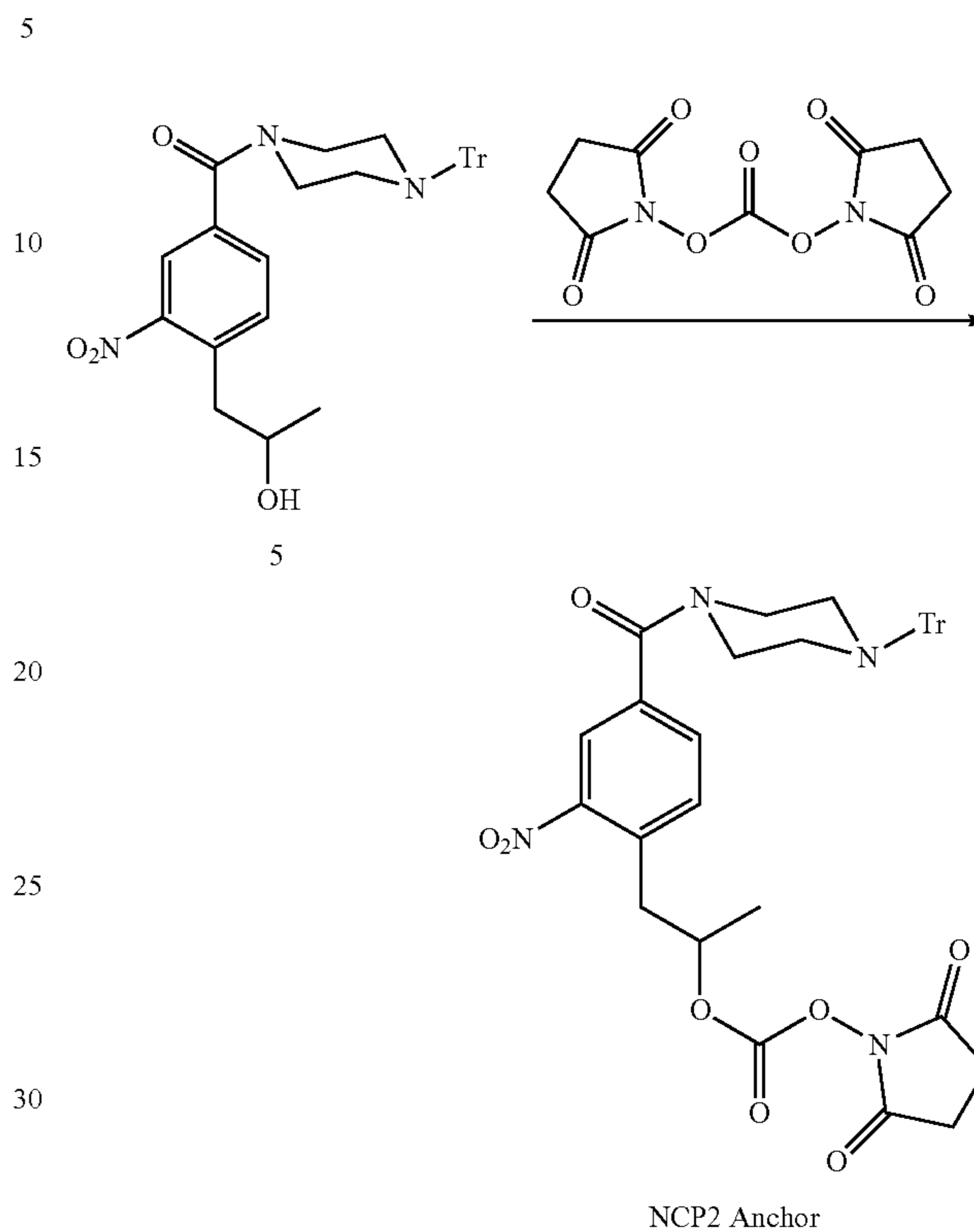
B. Preparation of (4-(2-Hydroxypropyl)-3-Nitrophenyl)(4-Triphenylpiperazin-1-yl)Methanone (5)



To a 100 L jacketed flask was charged under nitrogen 3600 g of 4 from the previous step and 9800 g THF. The stirred solution was cooled to $\leq 5^\circ$ C. The solution was diluted with 11525 g ethanol and 194 g of sodium borohydride was added over about 2 hours at 5° C. The reaction mixture was stirred an additional 2 hours at 5° C. The reaction was quenched with a solution of about 1.1 kg ammonium chloride in about 3 kg of water by slow addition to maintain the temperature at 10° C. The reaction mixture was stirred an additional 30 minutes, filtered to remove inorganics, and recharged to a 100 L jacketed flask and extracted with 23 kg of DCM. The organic layer was separated and the aqueous was twice more extracted with 4.7 kg of DCM each. The combined organic layers were washed with a solution of about 800 g of sodium chloride in about 3 kg of water, then dried over 2.7 kg of sodium sulfate. The suspension was filtered and the filter cake was washed with 2 kg of DCM. The combined filtrates were concentrated to 2.0 volumes, diluted with about 360 g of ethyl acetate, and evaporated. The crude product was loaded onto a silica gel column of 4 kg of silica packed with DCM under nitrogen and eluted with 2.3 kg ethyl acetate in 7.2 kg of DCM. The combined fractions were evaporated and the residue was taken up in 11.7 kg of toluene. The toluene solution was filtered and the filter cake was washed twice with 2 kg of toluene each. The filter cake was dried to a constant weight of 2.275 kg of compound 5 (46% yield from compound 3) HPLC 96.99%.

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5. Preparation of 2,5-Dioxopyrrolidin-1-yl(1-(2-Nitro-4-(4-triphenylmethylpiperazine-1-Carbonyl)Phenyl)Propan-2-yl) Carbonate (NCP2 Anchor)



To a 100 L jacketed flask was charged under nitrogen 4.3 kg of compound 5 (weight adjusted based on residual toluene by H^1 NMR; all reagents here after were scaled accordingly) and 12.7 kg pyridine. To this was charged 3.160 kg of DSC (78.91 weight % by H^1 NMR) while the internal temperature was maintained at $\leq 35^\circ$ C. The reaction mixture was aged for about 22 hours at ambience then filtered. The filter cake was washed with 200 g of pyridine. In two batches each comprising $\frac{1}{2}$ the filtrate volume, filtrate wash charged slowly to a 100 L jacketed flask containing a solution of about 11 kg of citric acid in about 50 kg of water and stirred for 30 minutes to allow for solid precipitation. The solid was collected with a filter funnel, washed twice with 4.3 kg of water per wash, and dried on the filter funnel under vacuum. The combined solids were charged to a 100 L jacketed flask and dissolved in 28 kg of DCM and washed with a solution of 900 g of potassium carbonate in 4.3 kg of water. After 1 hour, the layers were allowed to separate and the aqueous layer was removed. The organic layer was washed with 10 kg of water, separated, and dried over 3.5 kg of sodium sulfate. The DCM was filtered, evaporated, and dried under vacuum to 6.16 kg of NCP2 Anchor (114% yield).

Example 2: Anchor Loaded Resin Synthesis

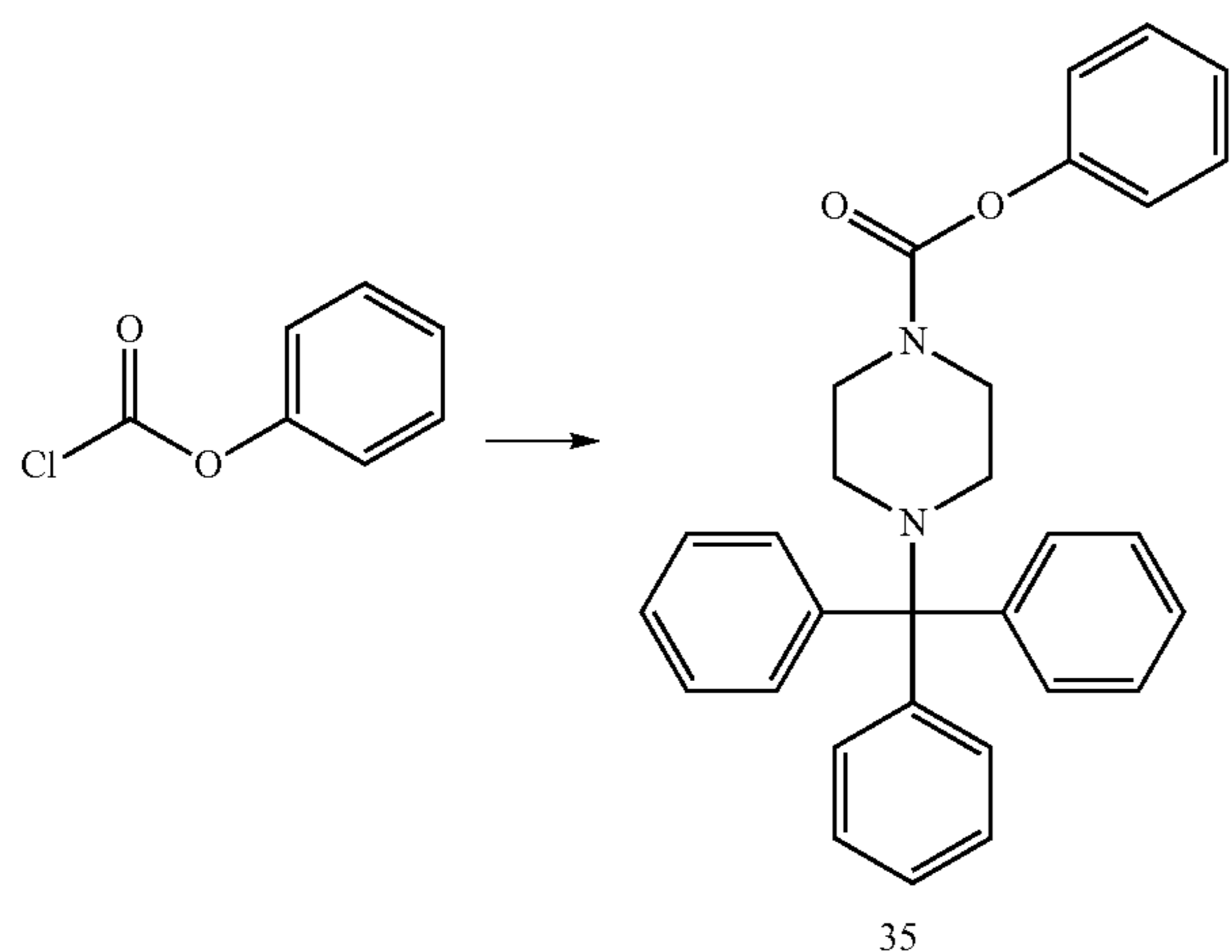
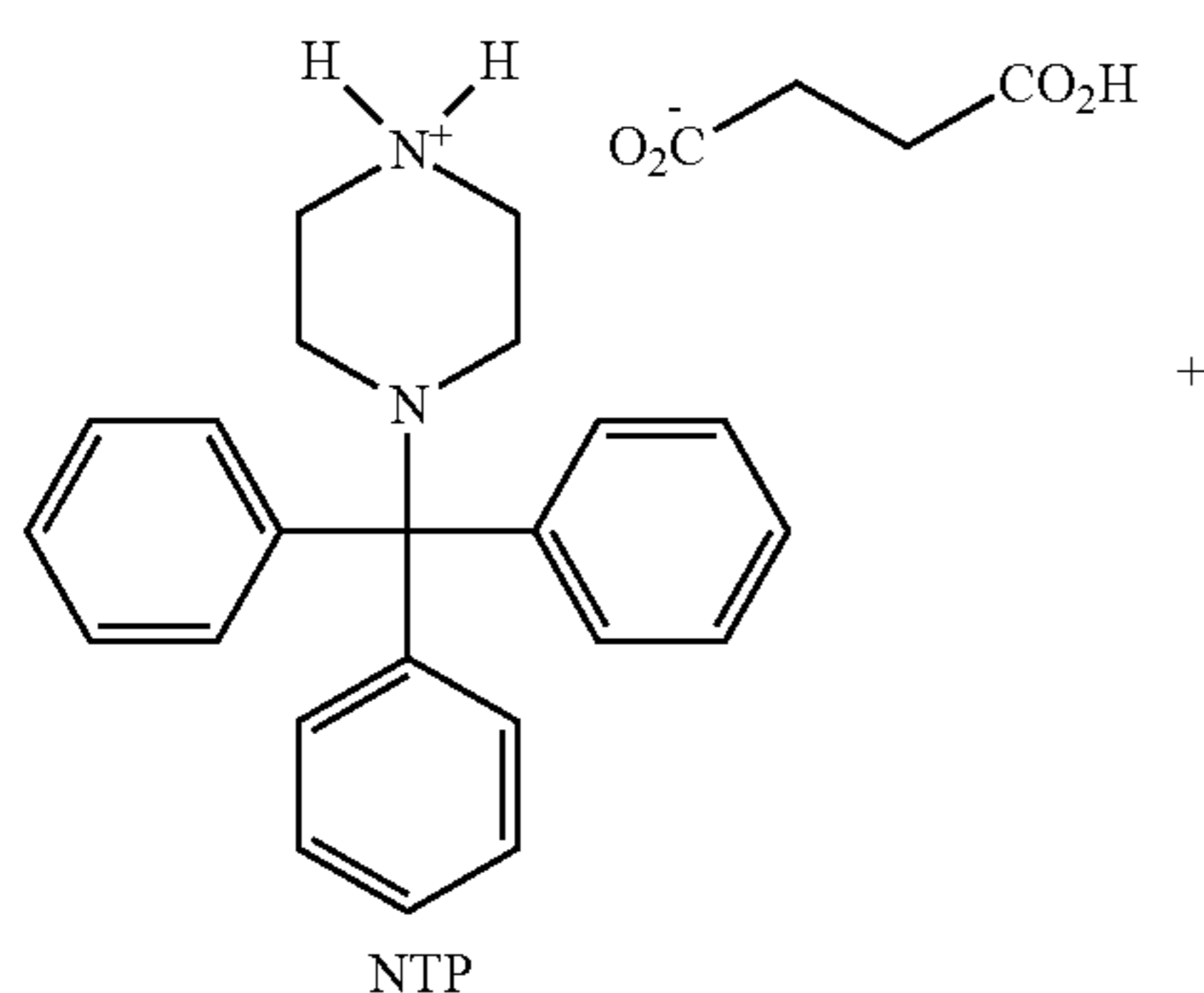
To a 75 L solid phase synthesis reactor with a Teflon stop cock was charged about 52 L of NMP and 2300 g of aminomethyl polystyrene resin. The resin was stirred in the NMP to swell for about 2 hours then drained. The resin was washed twice with about 4 L DCM per wash, then twice with 39 L Neutralization Solution per wash, then twice with 39 L

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of DCM per wash. The NCP2 Anchor Solution was slowly added to the stirring resin solution, stirred for 24 hours at room temperature, and drained. The resin was washed four times with 39 L of NMP per wash, and six times with 39 L of DCM per wash. The resin was treated and stirred with $\frac{1}{2}$ the DEDC Capping Solution for 30 minutes, drained, and was treated and stirred with the 2nd $\frac{1}{2}$ of the DEDC Capping Solution for 30 minutes and drained. The resin was washed six times with 39 L of DCM per wash then dried in an oven to constant weight of 3573.71 g of Anchor Loaded Resin.

Example 3: Preparation of Activated EG3 Tail (See FIG. 2)

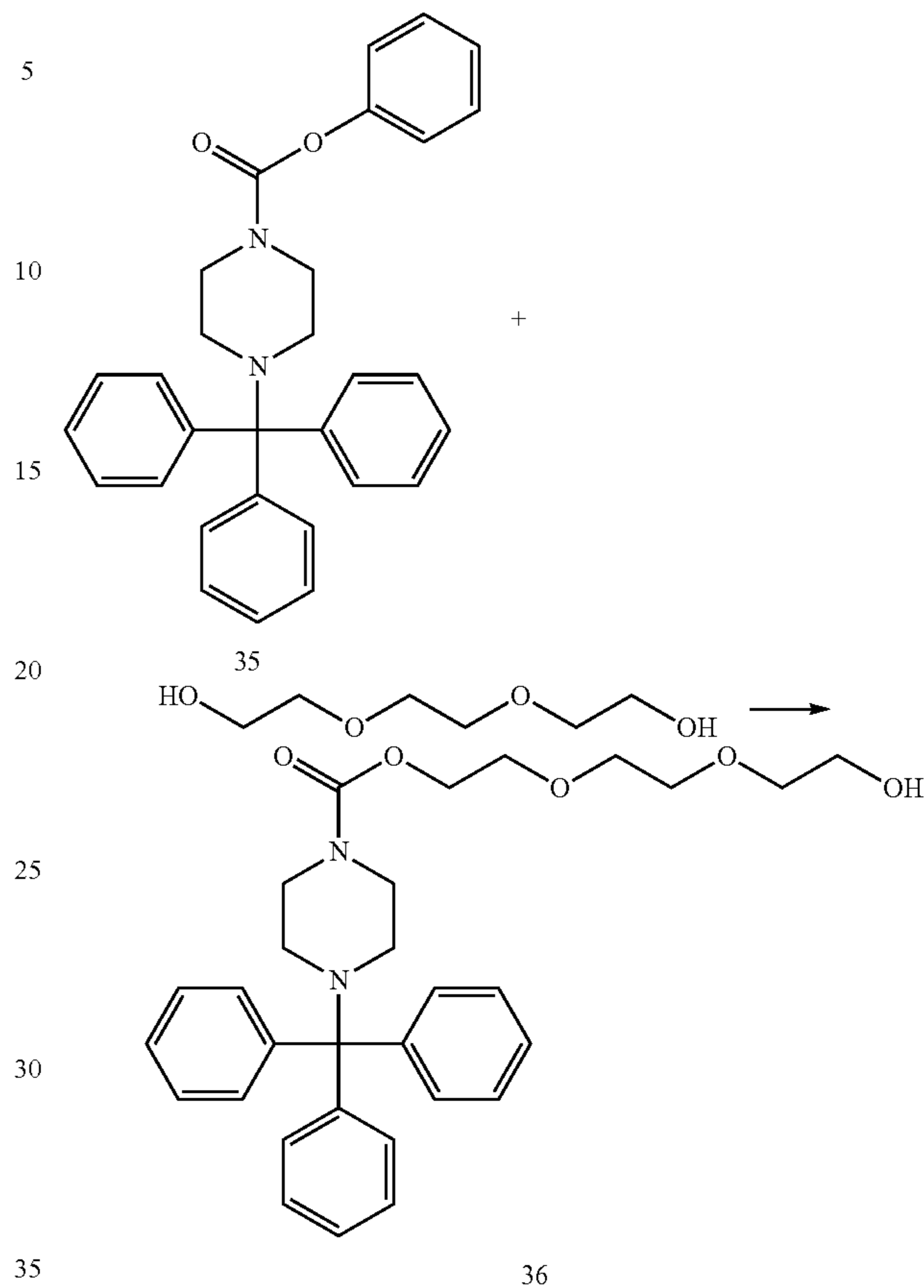
1. Preparation of Trityl Piperazine Phenyl Carbamate 35



To a cooled suspension of NTP in dichloromethane (6 mL/g NTP) was added a solution of potassium carbonate (3.2 eq) in water (4 mL/g potassium carbonate). To this two-phase mixture was slowly added a solution of phenyl chloroformate (1.03 eq) in dichloromethane (2 g/g phenyl chloroformate). The reaction mixture was warmed to 20° C. Upon reaction completion (1-2 hr), the layers were separated. The organic layer was washed with water, and dried over anhydrous potassium carbonate. The product 35 was isolated by crystallization from acetonitrile. Yield=80%.

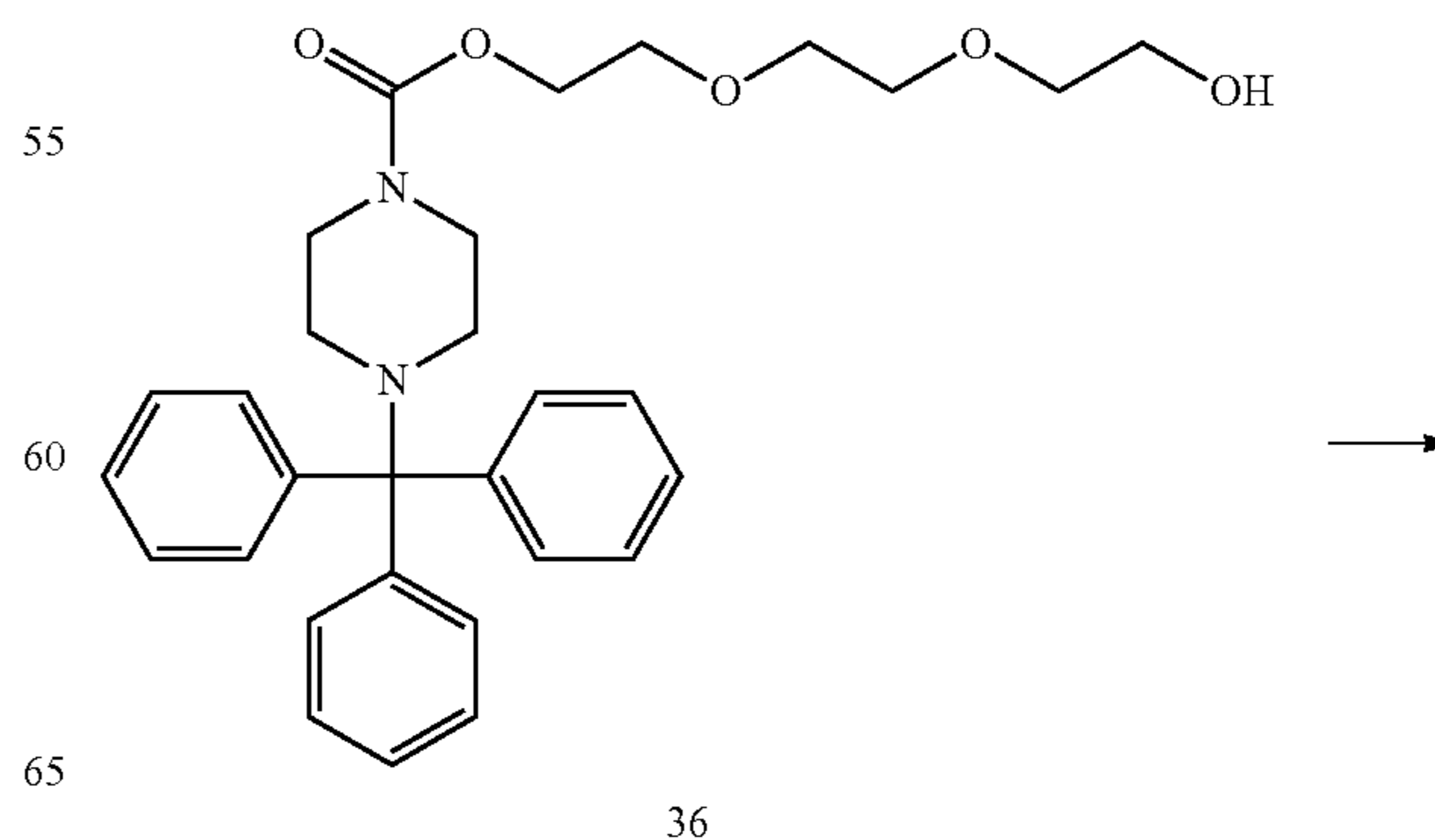
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2. Preparation of Carbamate Alcohol 36



Sodium hydride (1.2 eq) was suspended in 1-methyl-2-pyrrolidinone (32 mL/g sodium hydride). To this suspension were added triethylene glycol (10.0 eq) and compound 35 (1.0 eq). The resulting slurry was heated to 95° C. Upon reaction completion (1-2 hr), the mixture was cooled to 20° C. To this mixture was added 30% dichloromethane/methyl tert-butyl ether (v:v) and water. The product-containing organic layer was washed successively with aqueous NaOH, aqueous succinic acid, and saturated aqueous sodium chloride. The product 36 was isolated by crystallization from dichloromethane/methyl tert-butyl ether/heptane. Yield=90%.

3. Preparation of EG3 Tail Acid 37



Example 4: 50 L Solid-Phase Synthesis of
Eteplirsen [Oligomeric Compound (XII)] Crude
Drug Substance

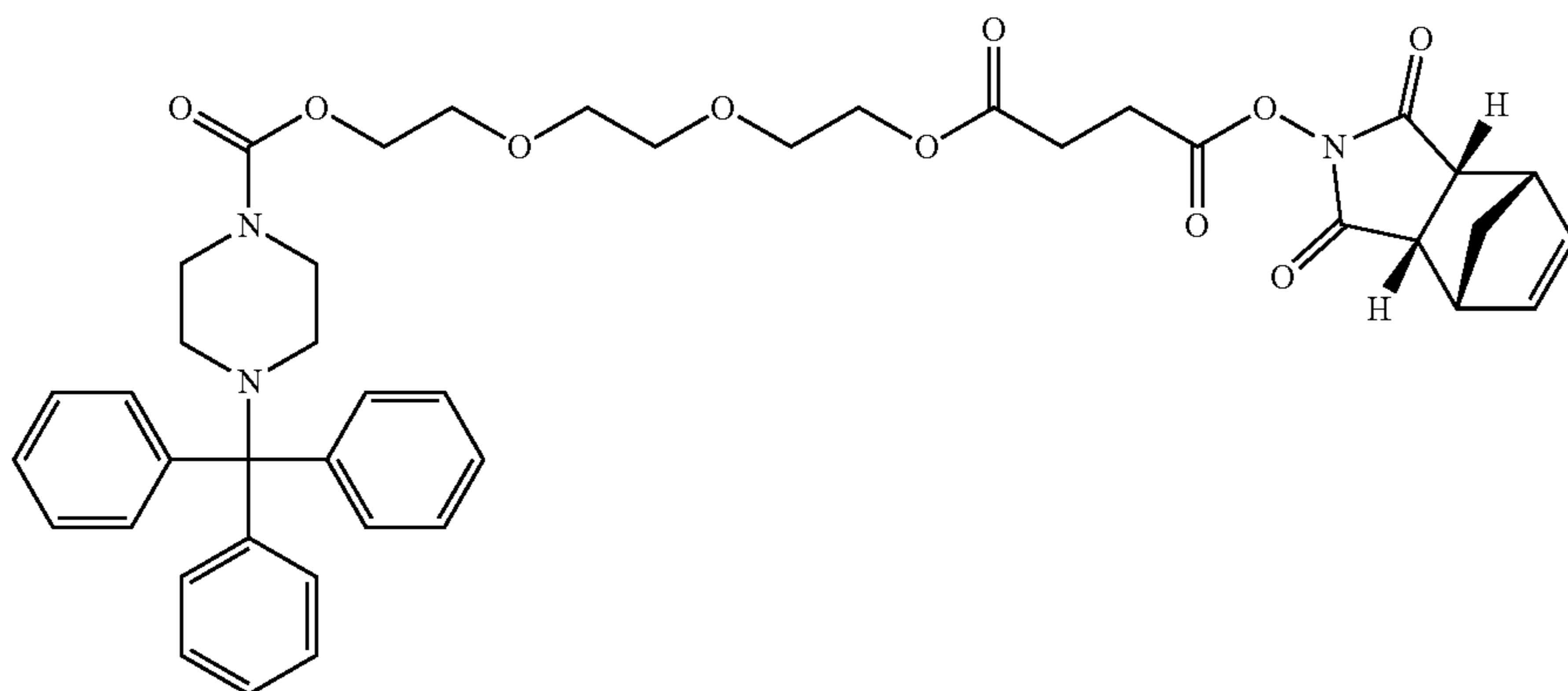
1. Materials

TABLE 2

Starting Materials				
Material Name	Chemical Name	CAS Number	Chemical Formula	MW
Activated A Subunit	Phosphoramidochloridic acid, N,N-dimethyl-,[6-[6- (benzoylamino)-9H-purin-9-yl]-4- (triphenylmethyl)-2- morpholinyl]methyl ester	1155373-30-0	C ₃₈ H ₃₇ ClN ₇ O ₄ P	722.2
Activated C Subunit	Phosphoramidochloridic acid, N,N-dimethyl-,[6-[4- (benzoylamino)-2-oxo-1(2H)- pyrimidinyl]-4-(triphenylmethyl)- 2-morpholinyl]methyl ester	1155373-31-1	C ₃₇ H ₃₇ ClN ₅ O ₅ P	698.2
Activated DPG Subunit	Propanoic Acid, 2,2-dimethyl-4- [[[9-[6- [[[chloro(dimethylamino)phosphin yl]oxy]methyl]-4- (triphenylmethyl)-2-morpholinyl]- 2-[(2-phenylacetyl)amino]-9H- purin-6-yl]oxy]methyl]phenyl ester	1155309-89-9	C ₅₁ H ₅₃ ClN ₇ O ₇ P	942.2
Activated T Subunit	Phosphoramidochloridic acid, N,N-dimethyl-,[6-(3,4-dihydro-5- methyl-2,4-dioxo-1(2H)- pyrimidinyl)]-4-(triphenylmethyl)- 2-morpholinyl]methyl ester	1155373-34-4	C ₃₁ H ₃₄ ClN ₄ O ₅ P	609.1
Activated EG3 Tail	Butanedioic acid, 1- [3aR,4S,7R,7aS)-1,3,3a,4,7,7a- hexahydro-1,3-dioxo-4,7-methano- 2H-isoindol-2-yl] 4-[2-[2-[2-[[[4- (triphenylmethyl)-1- piperazinyl]carbonyl]oxy]ethoxy]e thoxy]ethyl] ester	1380600-06-5	C ₄₃ H ₄₇ N ₃ O ₁₀	765.9

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Chemical Structures of Starting Materials:
A. Activated EG3 Tail



Compound (B)

L of CYTFA solution for 15 minutes and drained, and again washed with 5.5 L of CYTFA Solution for 15 minutes without draining to which 122 mL of 1:1 NEM/DCM was charged and the suspension stirred for 2 minutes and drained. The resin was washed twice with 5.5 L of Neutralization Solution for 5 minutes and drained, then twice with 5.5 L each of DCM and drained. A solution of 706.2 g of activated EG3 Tail (MW 765.85) and 234 mL of NEM in 3 L of DMI was charged to the resin and stirred for 3 hours at RT and drained. The resin was washed twice with 5.5 L each of Neutralization Solution for 5 minutes per each wash, and once with 5.5 L of DCM and drained. A solution of 374.8 g of benzoic anhydride and 195 mL NEM in 2680 mL NMP was charged and stirred for 15 minutes and drained. The resin was stirred with 5.5 L of Neutralization Solution for 5 minutes, then washed once with 5.5 L of DCM and twice with 5.5 L each of 30% TFE/DCM. The resin was suspended in 5.5 L of 30% TFE/DCM and held for 14 hours.

C. Subunit Coupling Cycles 1-30

i. Pre-Coupling Treatments

Prior to each coupling cycle as described in Table 4, the resin was: 1) washed with 30% TFE/DCM; 2) a) treated with CYTFA Solution 15 minutes and drained, and b) treated with CYTFA solution for 15 minutes to which was added 1:1 NEM/DCM, stirred, and drained; 3) stirred three times with Neutralization Solution; and 4) washed twice with DCM. See Table 4.

ii. Post Coupling Treatments

After each subunit solution was drained as described in Table 4, the resin was: 1) washed with DCM; and 2) washed

two times with 30% TFE/DCM. If the resin was held for a time period prior to the next coupling cycle, the second TFE/DCM wash was not drained and the resin was retained in said TFE/DCM wash solution. See Table 4.

iii. Activated Subunit Coupling Cycles

The coupling cycles were performed as described in Table 4.

iv. Final IPA Washing

After the final coupling step was performed as described in Table 4, the resin was washed 8 times with 19.5 L each of IPA, and dried under vacuum at room temperature for about 63.5 hours to a dried weight of 5,579.8 g.

D. Cleavage

The above resin bound Eteplirsén Crude Drug Substance was divided into two lots, each lot was treated as follows. A 2,789.9 g lot of resin was: 1) stirred with 10 L of NMP for 2 hrs, then the NMP was drained; 2) washed three times with 10 L each of 30% TFE/DCM; 3) treated with 10 L CYTFA Solution for 15 minutes; and 4) 10 L of CYTFA Solution for 15 minutes to which 130 ml 1:1 NEM/DCM was then added and stirred for 2 minutes and drained. The resin was treated three times with 10 L each of Neutralization Solution, washed six times with 10 L of DCM, and eight times with 10 L each of NMP. The resin was treated with a Cleaving Solution of 1530.4 g DTT and 2980 DBU in 6.96 L NMP for hours to detach the Eteplirsén Crude Drug Substance from the resin. The Cleaving solution was drained and retained in a separate vessel. The reactor and resin were washed with 4.97 L of NMP which was combined with the Cleaving Solution.

TABLE 4

Cycle No.: Subunit (SU)	Pre-coupling Treatment				Coupling Cycle		Post-Coupling Treatment	
	1	2	3	4	Quantity SU (g) NEM (L) DMI (L)	RT Coupling Time (Hrs.)	1	2
1:C	5.5 L	a) 5, 5 L b) 5.5 L, 122 ml	3 × 5.5 L	5.5 L	536.7 g; 195 ml NEM; 3.2 L DMI	5	5.5 L	2 × 5.5 L
2:T	7.0 L	a) 7 L b) 7 L, 158 ml	3 × 7 L	2 × 7 L	468.2 g and 195 ml NEM; 3.2 L DMI	4.25	7 L	2 × 7 L ²
3:C	8 L	a) 8 L b) 8 L, 182 ml	3 × 8 L	2 × 8 L	536.7 g; 195 ml NEM; 3.4 L DMI	4.25	8 L	2 × 8 L
4:C	9 L	a) 9 L b) 9 L, 206 ml	3 × 9 L	2 × 9 L	536.7 g; 195 ml NEM; 3.6 L DMI	4.25	9 L	2 × 9 L ³
5:A	9.5 L	a) 9.5 L b) 9.5 L, 220 ml	3 × 9.5 L	2 × 9.5 L	555.2 g; 195 ml NEM; 3.4 L DMI	4.25	9.5 L	2 × 9.5 L
6:A	10 L	a) 10 L b) 10 L, 232 ml	3 × 10 L	2 × 10 L	555.2 g; 195 ml NEM; 3.45 L DMI	4.25	10 L	2 × 10 L ⁴
7:C	11 L	a) 11 L b) 11 L, 256 ml	3 × 11 L	2 × 11 L	536.7 g; 195 ml NEM; 3.57 L DMI	4.25	11 L	2 × 11 L
8:A	11 L	a) 11 L b) 11 L, 256 ml	3 × 11 L	2 × 11 L	555.2 g; 195 ml NEM; 3.64 L DMI	4.25	11 L	2 × 11 L ⁵
9:T	11.5 L	a) 11.5 L b) 11.5 L 268 ml	3 × 11.5 L	2 × 11.5 L	468.2 g; 195 ml NEM; 3.72 L DMI	4.25	11.5 L	2 × 11.5 L
10:C	12 L	a) 12 L b) 12 L, 280 ml	3 × 12 L	2 × 12 L	536.7 g; 195 ml NEM; 3.96 L DMI	4.25	12 L	2 × 12 L ⁶
11:A	13.5 L	a) 13.5 L b) 13.5 L, 204 ml	3 × 13.5 L	2 × 13.5 L	721.7 g; 253 ml NEM; 4.02 L DMI	4.25	13.5 L	2 × 13.5 L
12:A	13.5 L	a) 13.5 L b) 13.5 L, 204 ml	3 × 13.5 L	2 × 13.5 L	721.7 g; 253 ml NEM; 4.02 L DMI	4.25	13.5 L	2 × 13.5 L ⁷
13:DPG	14 L	a) 14 L b) 14 L, 216 ml	3 × 14 L	2 × 14 L	941.9 g; 253 ml NEM; 4.02 L DMI	4.25	14 L	2 × 14 L
14:DPG	14.5 L	a) 14.5 L b) 14.5 L, 228 ml	3 × 14.5 L	2 × 14.5 L	941.9 g; 253 ml NEM; 4.1 L DMI	4.25	14.5 L	2 × 14.5 L ⁸
15:A	15.5 L	a) 15.5 L b) 15.5 L, 254 ml	3 × 15.5 L	2 × 15.5 L	721.7 g; 253 ml NEM; 4.26 L DMI	4.25	15.5 L	2 × 15.5 L
16:A	15.5 L	a) 15.5 L b) 15.5 L, 254 ml	3 × 15.5 L	2 × 15.5 L	721.7 g; 253 ml NEM; 4.26 L DMI	4.25	15.5 L	2 × 15.5 L ⁹

TABLE 4-continued

Cycle No.:	Pre-coupling Treatment				Coupling Cycle		Post-Coupling Treatment	
	1	2	3	4	Quantity SU (g)	RT	1	2
Subunit (SU)	TFE/DCM Wash	CYTFA Solution ¹	Neutralization Solution	DCM Wash	NEM (L) DMI (L)	Coupling Time (Hrs.)	DCM Wash	TFE/DCM Wash
17:DPG	16 L	a) 16 L b) 16 L, 366 ml	3 × 16 L	2 × 16 L	941.9 g; 253 ml NEM; 4.4 L DMI	4.75	16 L	2 × 16 L
18:A	16.5 L	a) 16.5 L b) 16.5 L, 378 ml	3 × 16.5 L	2 × 16.5 L	721.7 g; 253 ml NEM; 4.4 L DMI	4.25	16.5 L	2 × 16.5 L ¹⁰
19:T	16.5 L	a) 16.5 L b) 16.5 L, 378 ml	3 × 16.5 L	2 × 16.5 L	608.7 g; 253 ml NEM; 4.57 L DMI	4.25	16.5 L	2 × 16.5 L
20:DPG	17 L	a) 17 L b) 17 L, 390 ml	3 × 17 L	2 × 17 L	941.9 g; 253 ml NEM; 4.57 L DMI	4.75	17 L	2 × 17 L ¹¹
21:DPG	17 L	a) 17 L b) 17 L, 390 ml	3 × 17 L	2 × 17 L	1159.2 g; 311 ml NEM; 4.72 L DMI	4.25	17 L	2 × 17 L
22:C	17.5 L	a) 17.5 L b) 17.5 L, 402 ml	3 × 17.5 L	2 × 17.5 L	858.7 g; 311 ml NEM; 4.72 L DMI	4.75	17.5 L	2 × 17.5 L ¹²
23:A	17.5 L	a) 17.5 L b) 17.5 L, 402 ml	3 × 17.5 L	2 × 17.5 L	888.3 g; 311 ml NEM; 4.88 L DMI	4.25	17.5 L	2 × 17.5 L
24:T	18 L	a) 18 L b) 18 L, 414 ml	3 × 18 L	2 × 18 L	749.1 g; 311 ml NEM; 4.95 L DMI	4.25	18 L	2 × 18 L ¹³
25:T	18 L	a) 18 L b) 18 L, 414 ml	3 × 18 L	2 × 18 L	749.1 g; 311 ml NEM; 5.1 L DMI	4.25	18 L	2 × 18 L
26:T	18.5 L	a) 18.5 L b) 18.5 L, 426 ml	3 × 18.5 L	2 × 18.5 L	749.1 g; 311 ml NEM; 5.1 L DMI	4.25	18.5 L	2 × 18.5 L ¹⁴
27:C	18.5 L	a) 18.5 L b) 18.5 L, 426 ml	3 × 18.5 L	2 × 18.5 L	858.7 g; 311 ml NEM; 5.25 L DMI	4.25	18.5 L	2 × 18.5 L
28:T	19 L	a) 19 L b) 19 L, 438 ml	3 × 19 L	2 × 19 L	749.1 g; 311 ml NEM; 5.25 L DMI	4.25	19 L	2 × 19 L ¹⁵
29:A	19 L	a) 19 L b) 19 L, 438 ml	3 × 19 L	2 × 19 L	888.3 g; 311 ml NEM; 5.41 L DMI	4.25	19 L	2 × 19 L
30:DPG	19.5 L	a) 19.5 L b) 19.5 L, 450 ml	3 × 19.5 L	2 × 19.5 L	1159.2 g; 311 ml NEM; 5.44 L DMI	4.75	19.5 L	2 × 19.5 L

¹ml indicates the amount of 1:1 NEM/DCM²Resin held at this step for ½ day³Resin held at this step for ½ day⁴Resin held at this stage for 0.4 days⁵Resin held at this stage for 2.5 days⁶Resin held at this stage for ½ day⁷Resin held at this stage for 0.4 days⁸Resin held at this stage for 0.4 days⁹Resin held at this stage for 0.4 days¹⁰Resin held at this stage for 1.5 days¹¹Resin held at this stage for 0.3 days¹²Resin held at this stage for 0.4 days¹³Resin held at this stage for 0.4 days¹⁴Resin held at this stage for 0.4 days¹⁵Resin held at this stage for 0.3 days

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E. Deprotection

The combined Cleaving Solution and NMP wash were transferred to a pressure vessel to which was added 39.8 L of NH₄H (NH₃·H₂O) that had been chilled to a temperature of -10° to -25° C. in a freezer. The pressure vessel was sealed and heated to 45° C. for 16 hrs then allowed to cool to 25° C. This deprotection solution containing the Etep-lirsén crude drug substance was diluted 3:1 with purified water and pH adjusted to 3.0 with 2M phosphoric acid, then to pH 8.03 with NH₄OH. HPLC (C18) 73-74% (FIG. 1).

TABLE 5

Data of Figure 1						
Peak #	Compound Name	Retention Time (min)	Rel. Ret. Product	Area {mAu*min}	Rel. Area %	Plates (USP)
1		2.488	0.381	0.821928	0.18	1105
2		3.047	0.467	17.661449	3.91	4047

TABLE 5-continued

Data of Figure 1						
Peak #	Compound Name	Retention Time (min)	Rel. Ret. Product	Area {mAu*min}	Rel. Area %	Plates (USP)
3		3.324	0.509	0.818258	0.18	n.a.
4		3.605	0.552	0.465598	0.10	7
5		4.213	0.645	6.558899	1.45	301
6		4.504	0.690	3.324238	0.74	191690
7		5.160	0.790	5.644073	1.25	651
8	AVI-4658	6.531	1.000	332.238891	73.47	2313
9		7.269	1.113	2.063159	0.46	n.a.
10		7.643	1.170	5.556411	1.23	2734
11		8.139	1.246	8.711530	1.93	3572
12		8.382	1.283	4.654783	1.03	1835
13		8.678	1.329	0.562426	0.12	n.a.
14		9.009	1.379	12.031923	2.66	6078
15		9.500	1.455	0.385563	0.09	n.a.
16		9.626	1.474	1.171507	0.26	46084
17		9.898	1.516	0.484362	0.11	21328
18		10.598	1.623	14.589918	3.23	n.a.

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TABLE 5-continued

Data of Figure 1						
Peak #	Compound Name	Retention Time (min)	Rel. Ret. Time. Product	Area {mAu*min}	Rel. Area %	Plates (USP)
19		10.680	1.635	7.520577	1.66	918
20		10.811	1.656	8.604558	1.90	296
21		11.045	1.691	18.351689	4.06	49919

Example 5: Purification of Eteplirsen Crude Drug Substance

The deprotection solution from Example 2, part D, containing the Eteplirsen crude drug substance was loaded onto a column of ToyoPearl Super-Q 650S anion exchange resin (Tosoh Bioscience) and eluted with a gradient of 0-35% B over 17 column volume (Buffer A: 10 mM sodium hydroxide; Buffer B: 1 M sodium chloride in 10 mM sodium hydroxide) and fractions of acceptable purity (C18 and SCX HPLC) were pooled to a purified drug product solution. HPLC (FIG. 2): 97.74% (C18) 94.58% (SCX).

The purified drug substance solution was desalted and lyophilized to 1959 g purified Eteplirsen drug substance. Yield 61.4%; HPLC (FIG. 3): 97.7% (C18) 94.6% (SCX).

TABLE 6

Data of Figure 2						
Peak #	Compound Name	Retention Time (min)	Rel. Ret. Time. (Product)	Area {mAu*min}	Area Percent	Plates (USP)
1		6.837	0.750	0.050757	0.058	41574
2		7.405	0.813	0.303271	0.344	841
3		8.086	0.887	1.130007	1.280	13
4		8.615	0.946	2.265128	2.567	761
5	AVI-4658	9.111	1.000	83.468700	94.583	4405
6		10.019	1.100	0.704599	0.798	n.a.
7		11.069	1.215	0.326550	0.370	3044

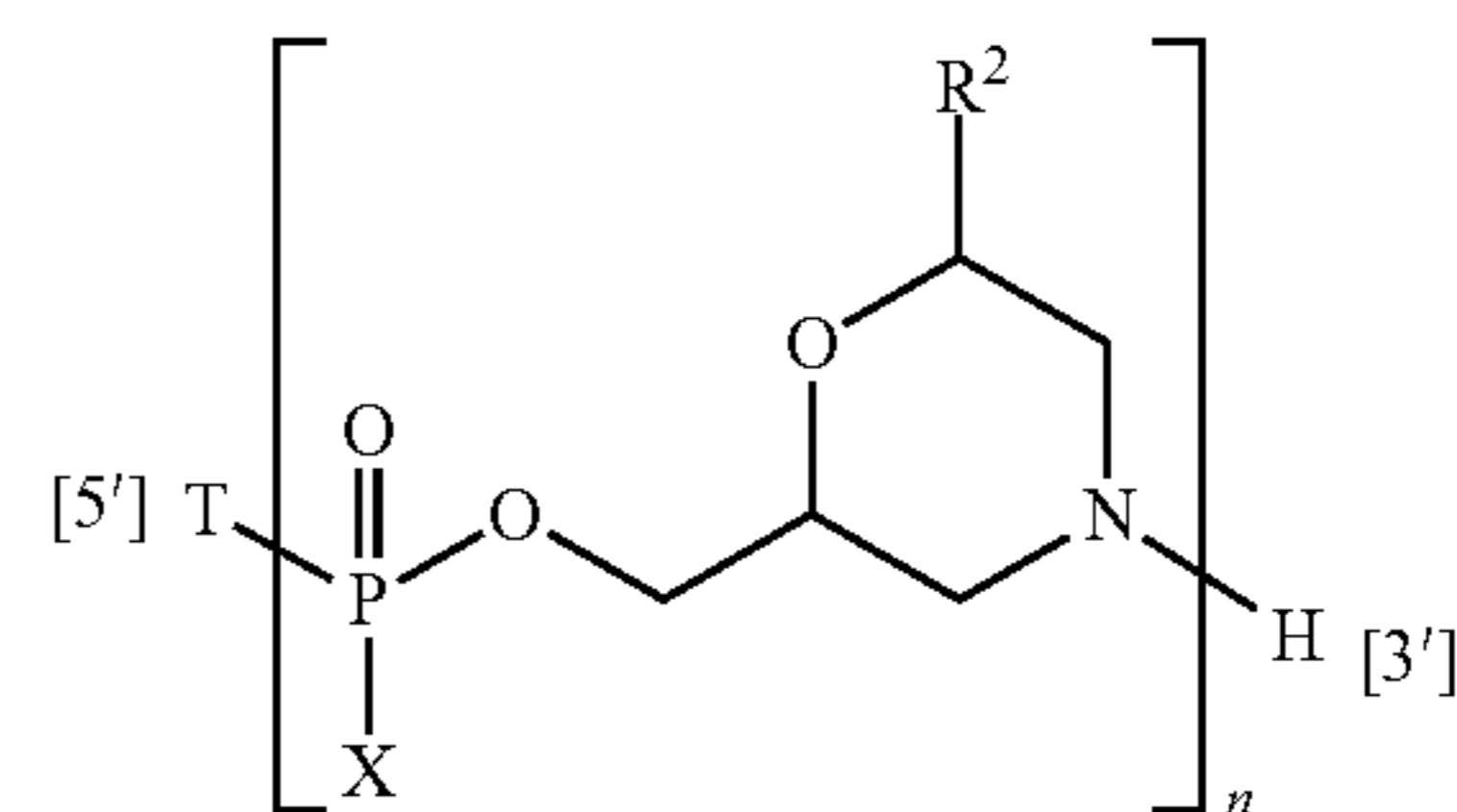
TABLE 7

Data of Figure 3						
Peak #	Compound Name	Retention Time (min)	Rel. Ret. Time. (Product)	Area {mAu*min}	Area Percent	Plates (USP)
1		6.866	0.751	0.044399	0.063	608
2		7.794	0.852	0.280589	0.397	n.a.
3		8.188	0.895	0.816793	1.156	209
4		8.644	0.945	1.842896	2.608	1147
5	AVI-4658	9.145	1.000	66.857088	94.622	4664
6		10.058	1.100	0.575793	0.815	n.a.
7		11.103	1.214	0.239454	0.339	4375

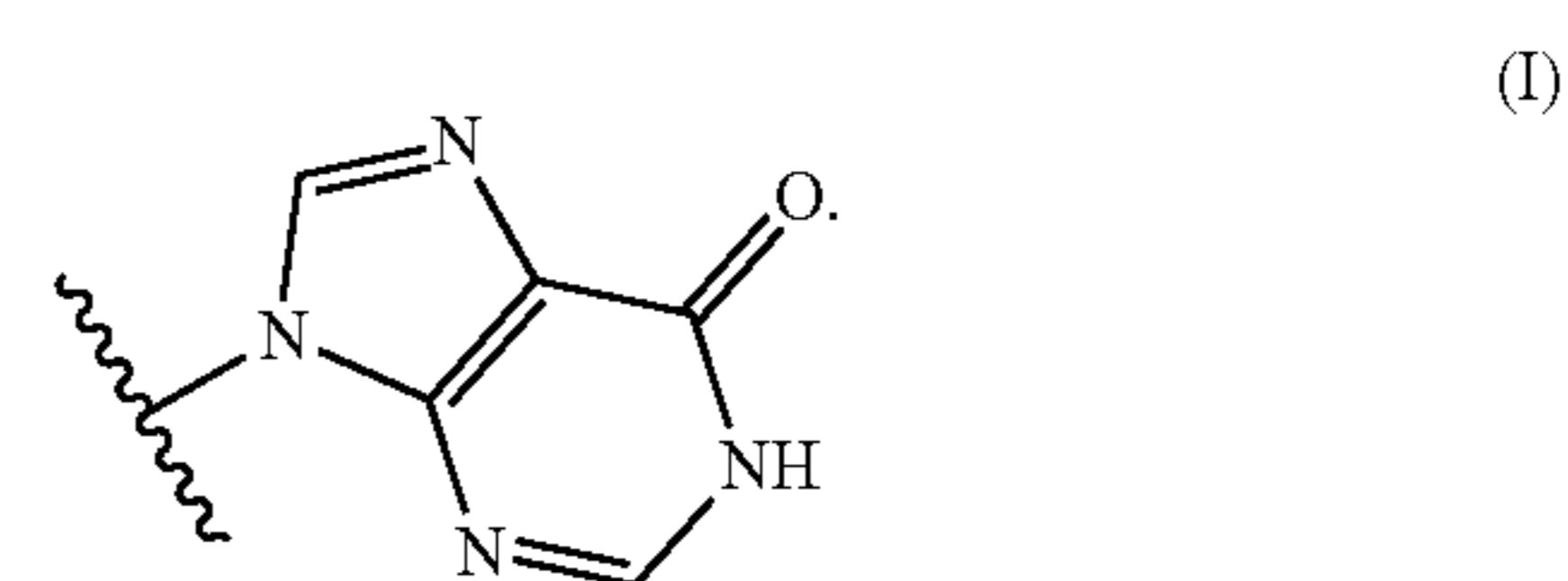
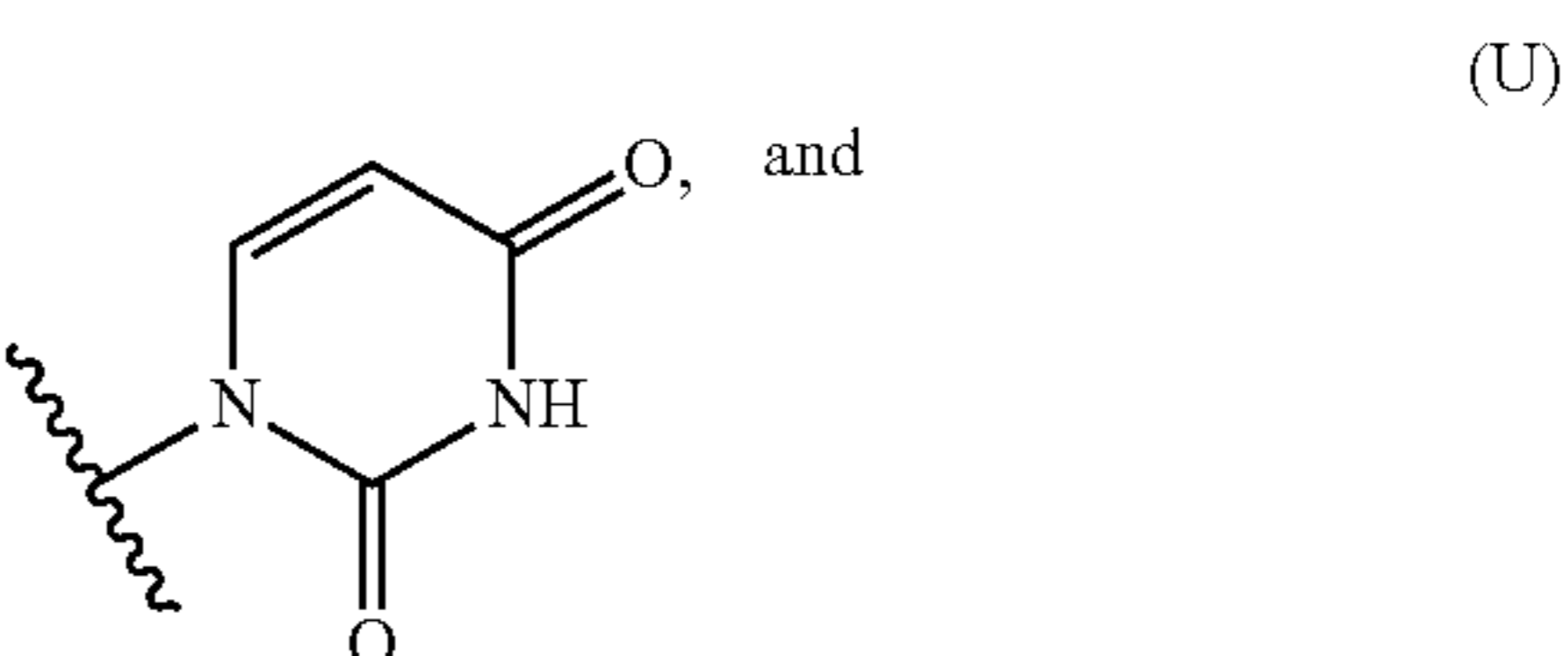
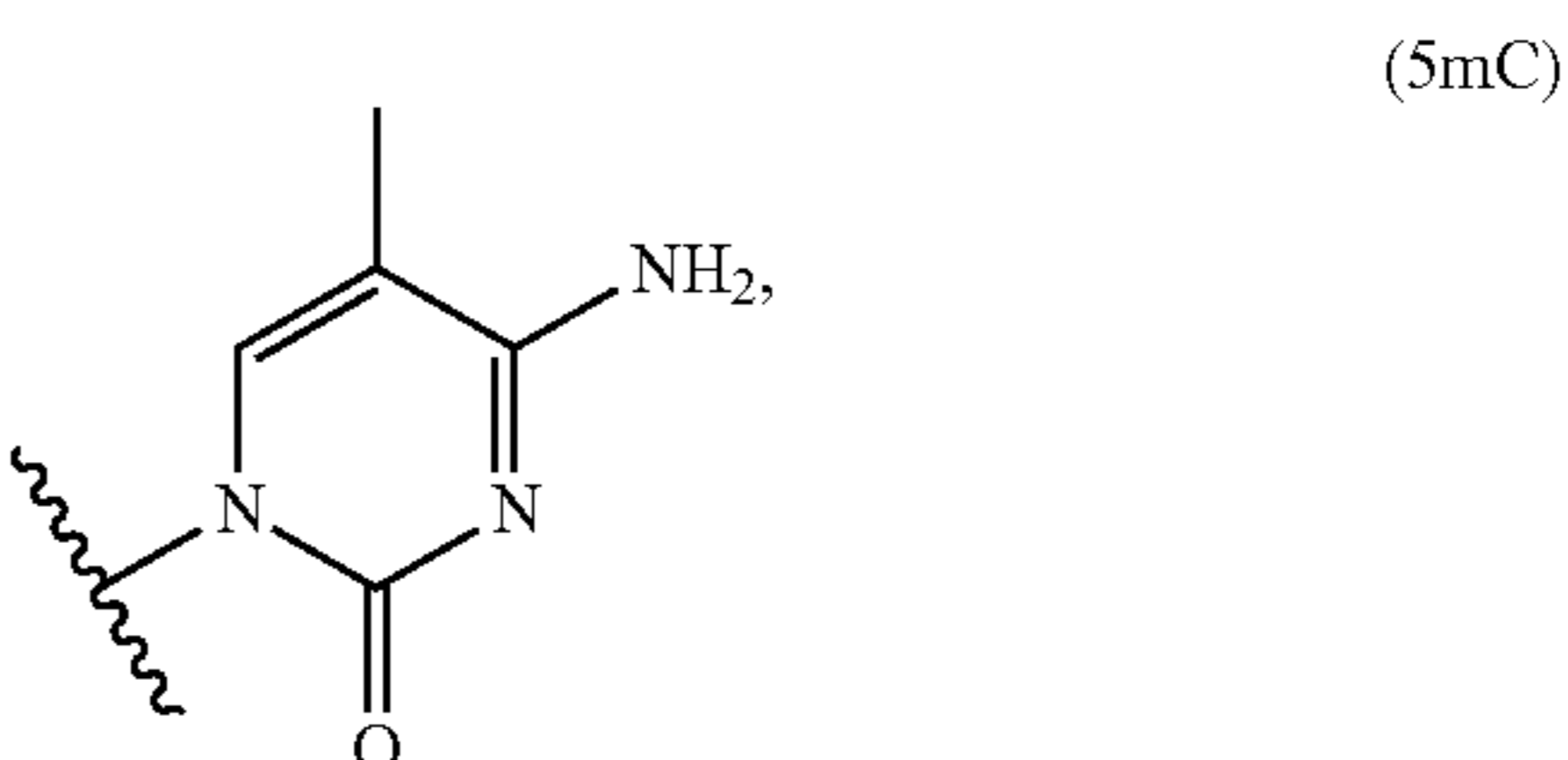
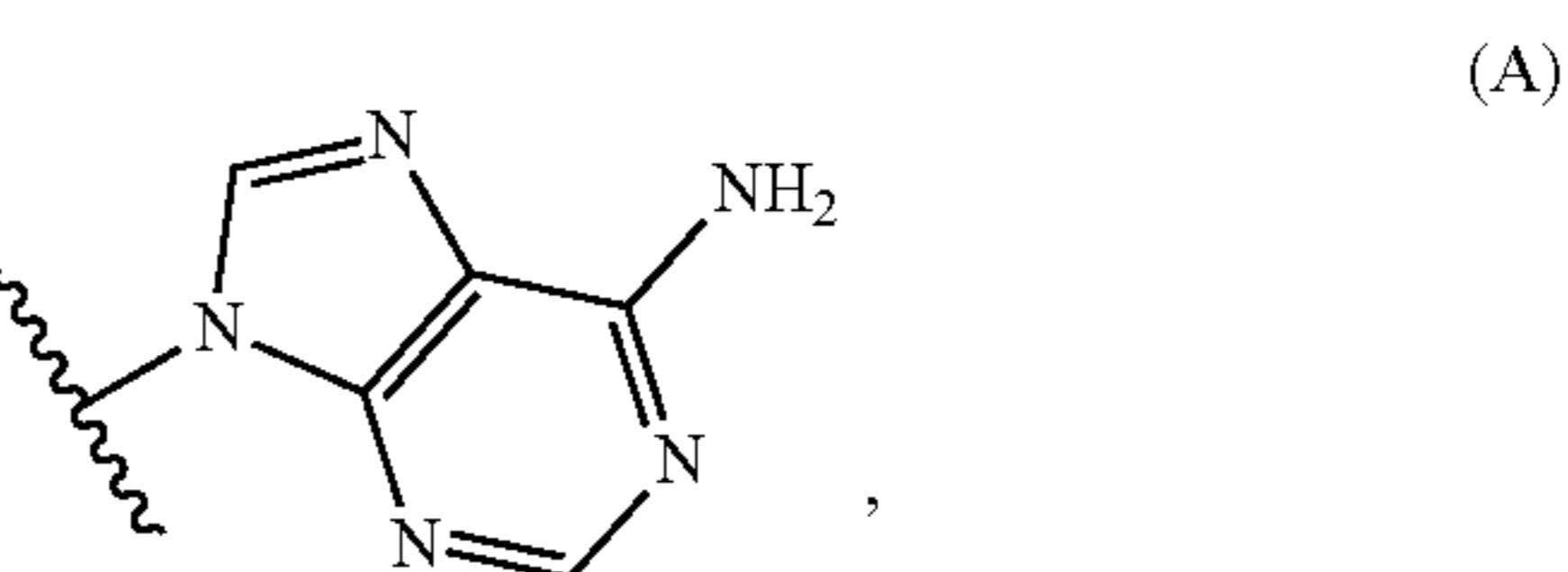
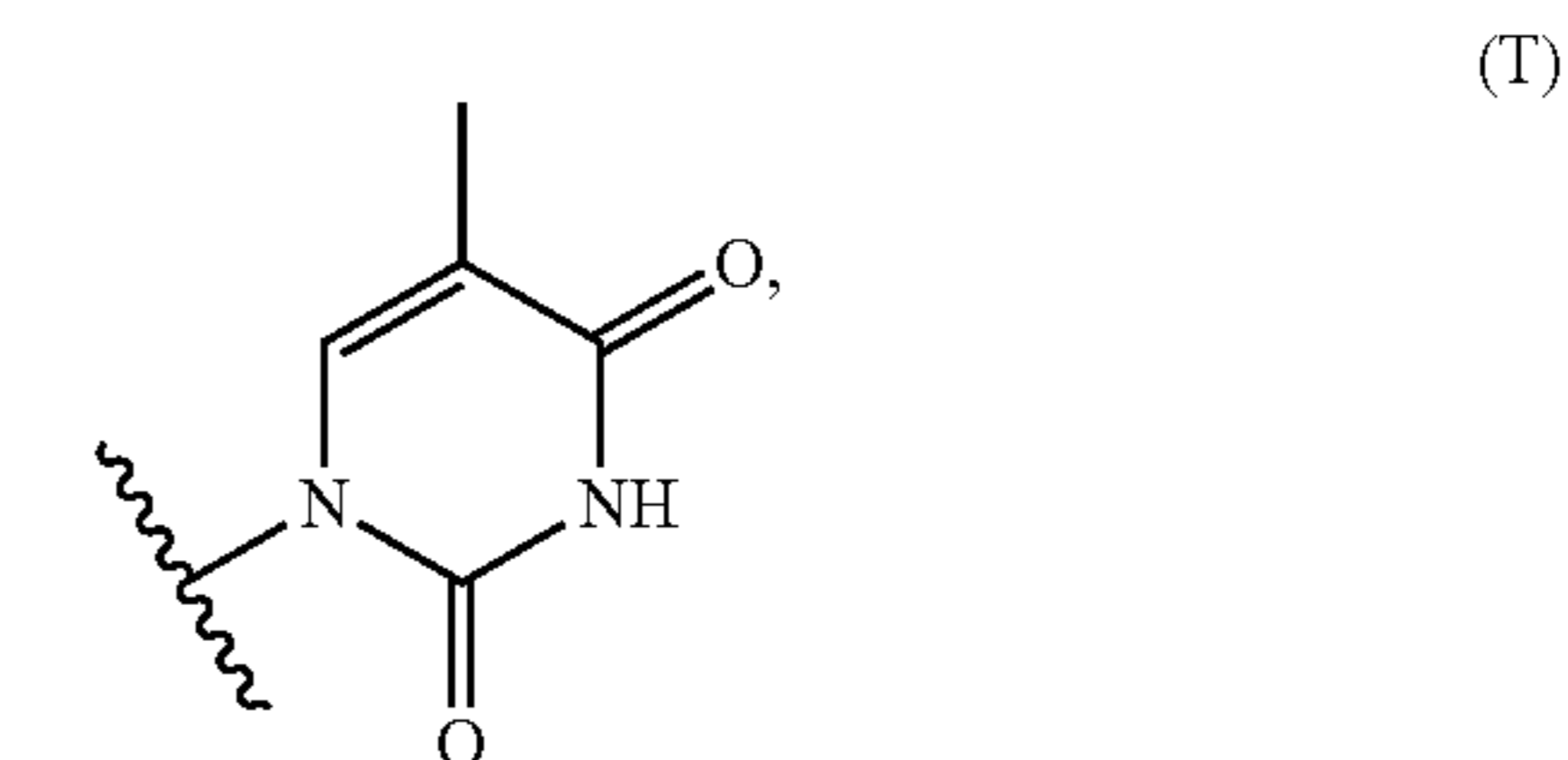
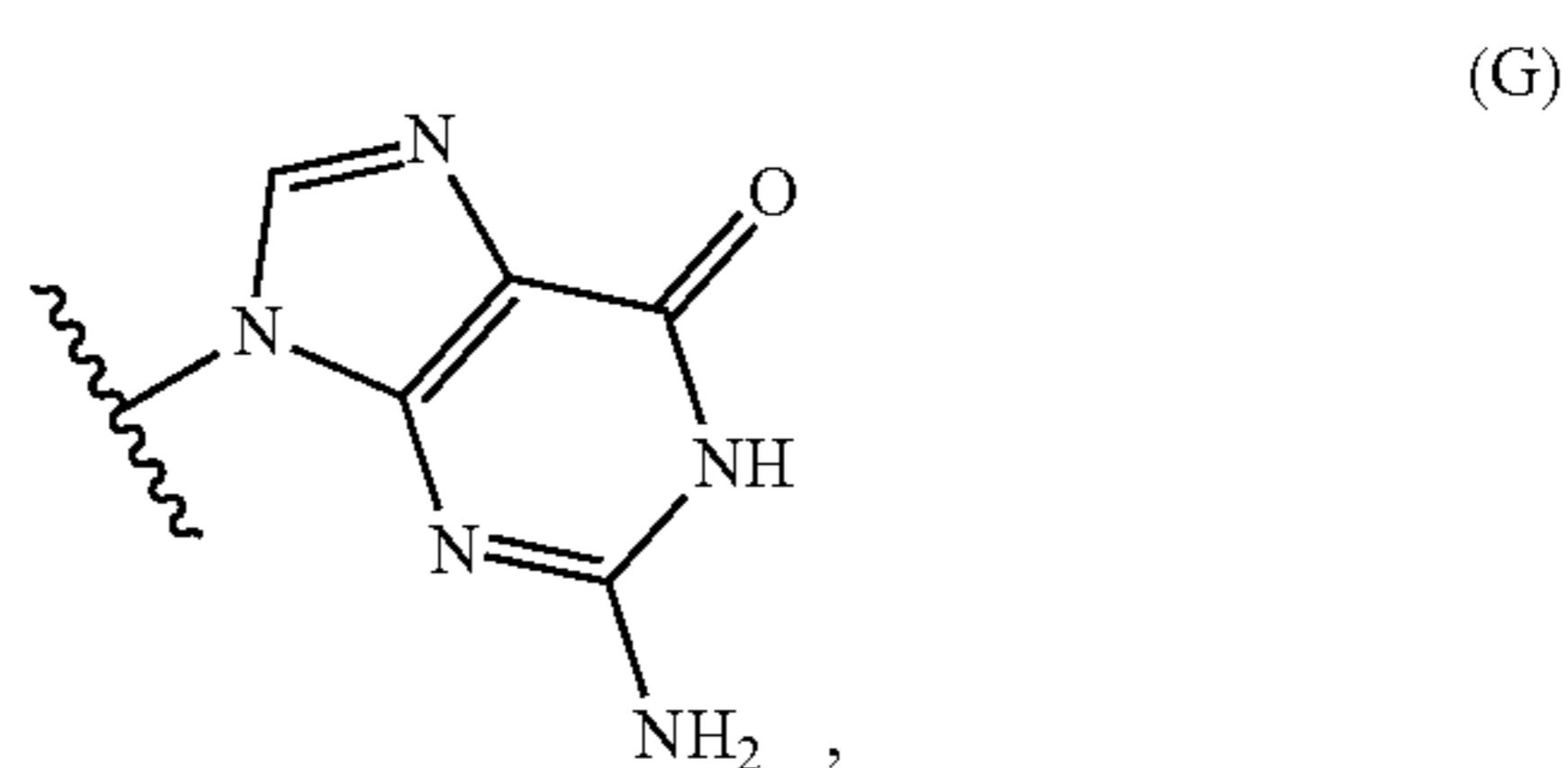
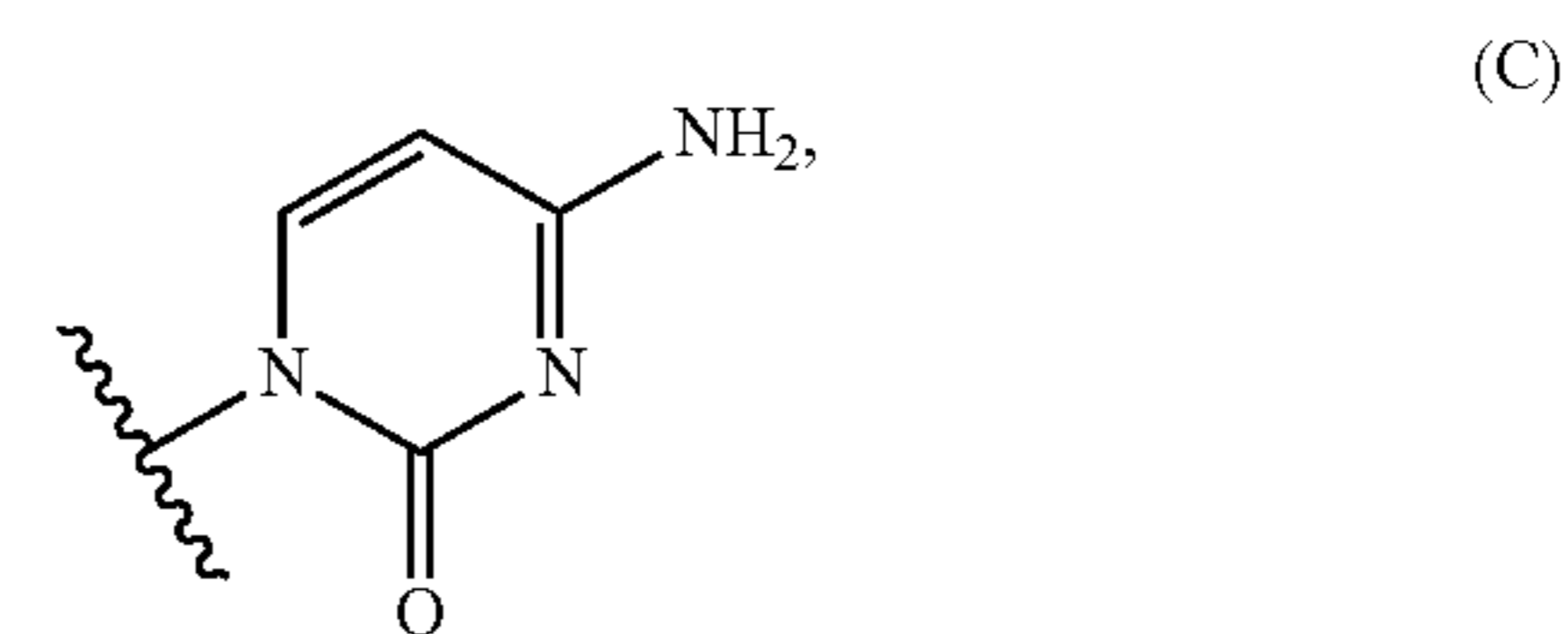
Example 6: Additional Oligomers Prepared by the Process of the Disclosure

The following oligomers of the following sequences were prepared according to the methods of Examples 4 and 5 about:

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where R² is, at each occurrence, independently selected from the group consisting of:

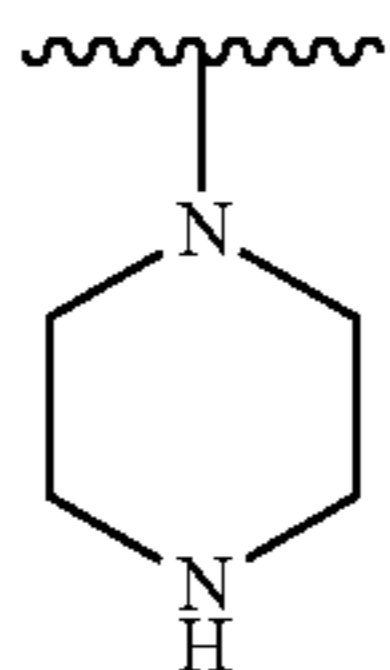


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TABLE 8

Oligomer R ² at each position from 1 to n and 5' to 3'			
Name	n	R ² 5' to 3' and 1 to n	T
PMO1	22	CAA TGC CAT CCT GGA GTT CCT G (SEQ ID NO: 3)	EG3
PMO2	25	GTT GCC TCC GGT TCT GAA GGT GTT C (SEQ ID NO: 4)	EG3
AVI-7100	20	CGGT + TAGAAGAC + TCATC + TTT (SEQ ID NO: 5)	EG3
AVI-7537	19	GCC + ATGGT + TTT + TTC + TC + AGG (SEQ ID NO: 6)	EG3
AVI-7288	23	GAATATTAAC + AI + AC + TGAC + A AGTC (SEQ ID NO: 7)	EG3

In the above Table 8, "+" indicates wherein X is



otherwise X is —N(CH₃)₂.

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TABLE 9

Acronyms	
Acronym	Name
DBU	1,8-Diazabicycloundec-7-ene
DCM	Dichloromethane
DIPEA	N,N-Diisopropylethylamine
DMI	1,3-Dimethyl-2-imidazolidinone
DTT	Dithiothreitol
IPA	Isopropyl alcohol
MW	Molecular weight
NEM	N-Ethylmorpholine
NMP	N-Methyl-2-pyrrolidone
RT	Room temperature
TFA	2,2,2-Trifluoroacetic acid
TFE	2,2,2-Trifluoroethanol

INCORPORATION BY REFERENCE

The contents of all references (including literature references, issued patents, published patent applications, and co-pending patent applications) cited throughout this application are hereby expressly incorporated herein in their entireties. Unless otherwise defined, all technical and scientific terms used herein are accorded the meaning commonly known to one with ordinary skill in the art.

EQUIVALENTS

Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, many equivalents of the specific embodiments of the disclosure described herein. Such equivalents are intended to be encompassed by the following claims.

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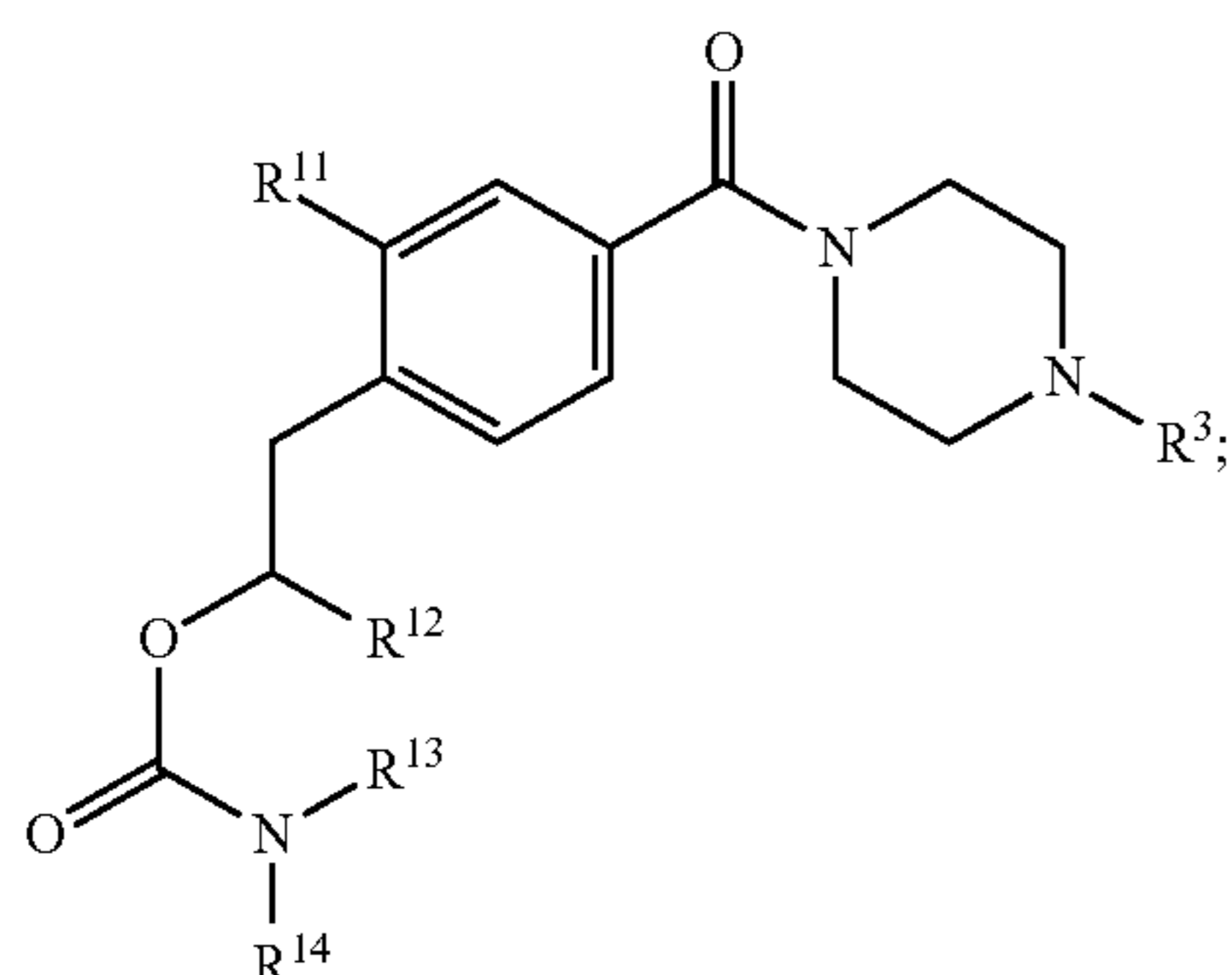
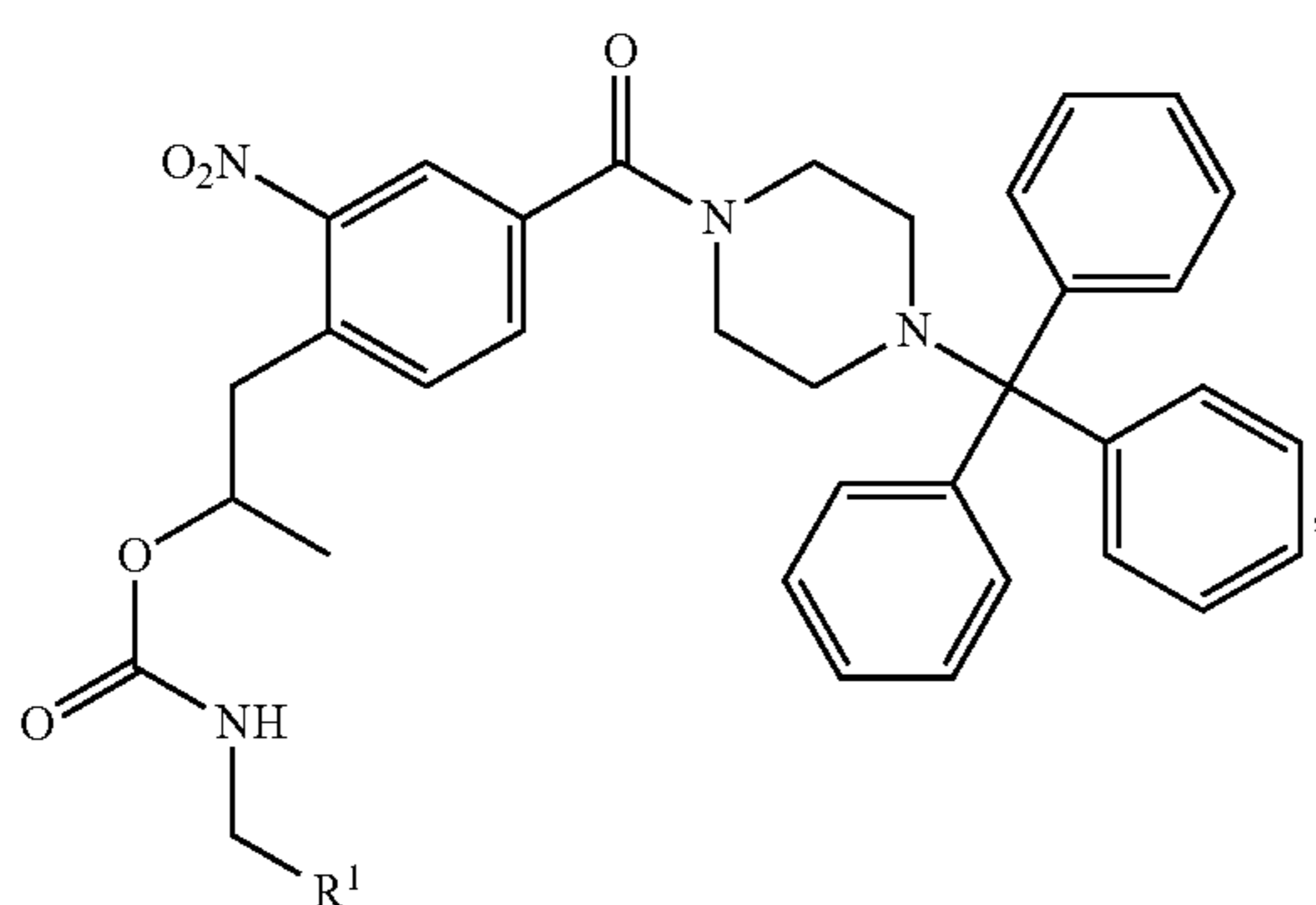
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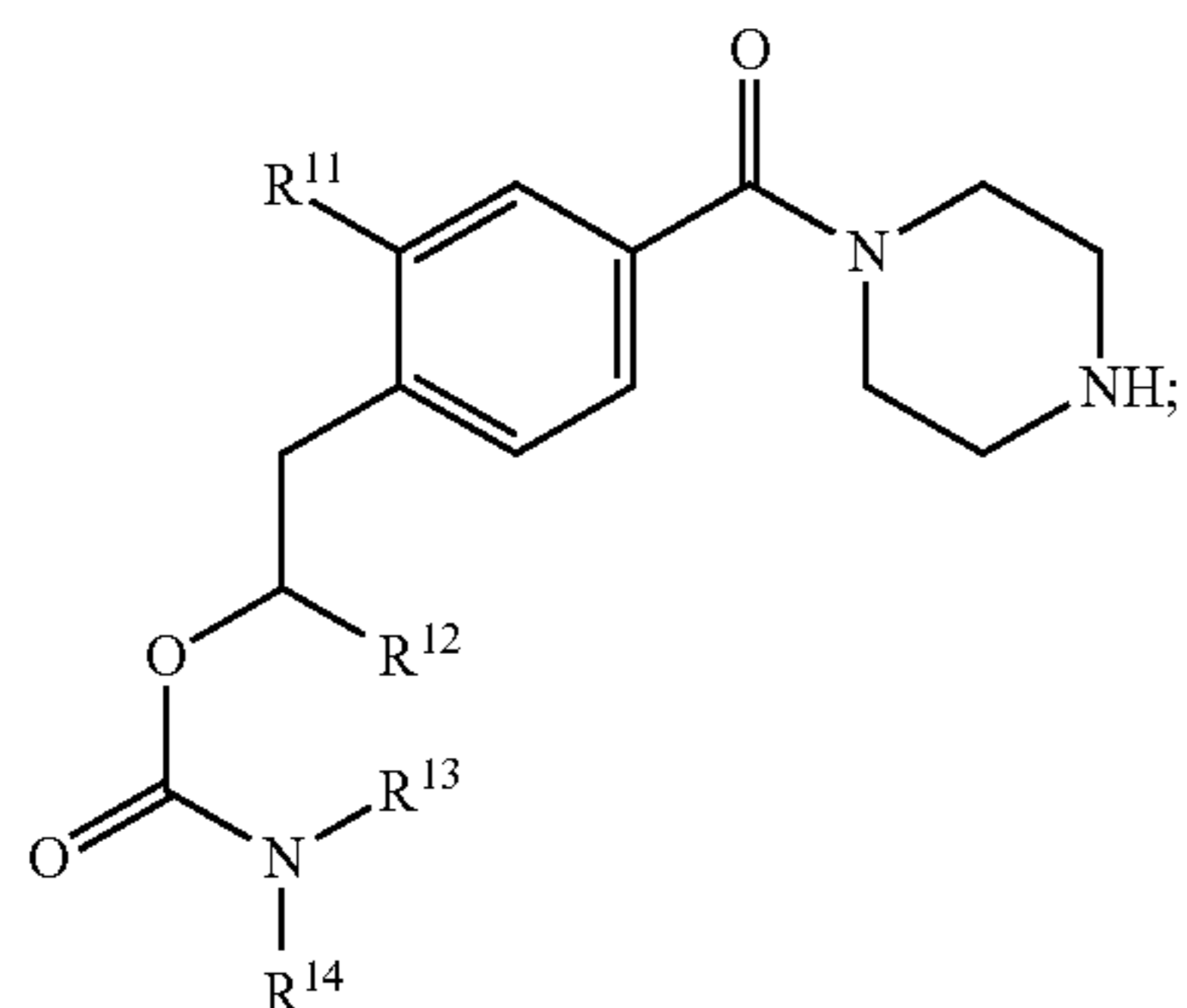
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The invention claimed is:

1. A compound of Formula (B1):

or a pharmaceutically acceptable salt thereof
whereinR¹¹ is selected from the group consisting of halo, CN,
and NO₂;R¹² is C₁₋₆-alkyl;R¹³ is H or C₁₋₆-alkyl; andR¹⁴ is a first linker bound to a support-medium,R³ is selected from the group consisting of trityl,
monomethoxytrityl, dimethoxytrityl and
trimethoxytrityl.2. The compound of claim 1, wherein the compound of
Formula (B1) is of Formula (I):wherein R¹ is a support-medium.

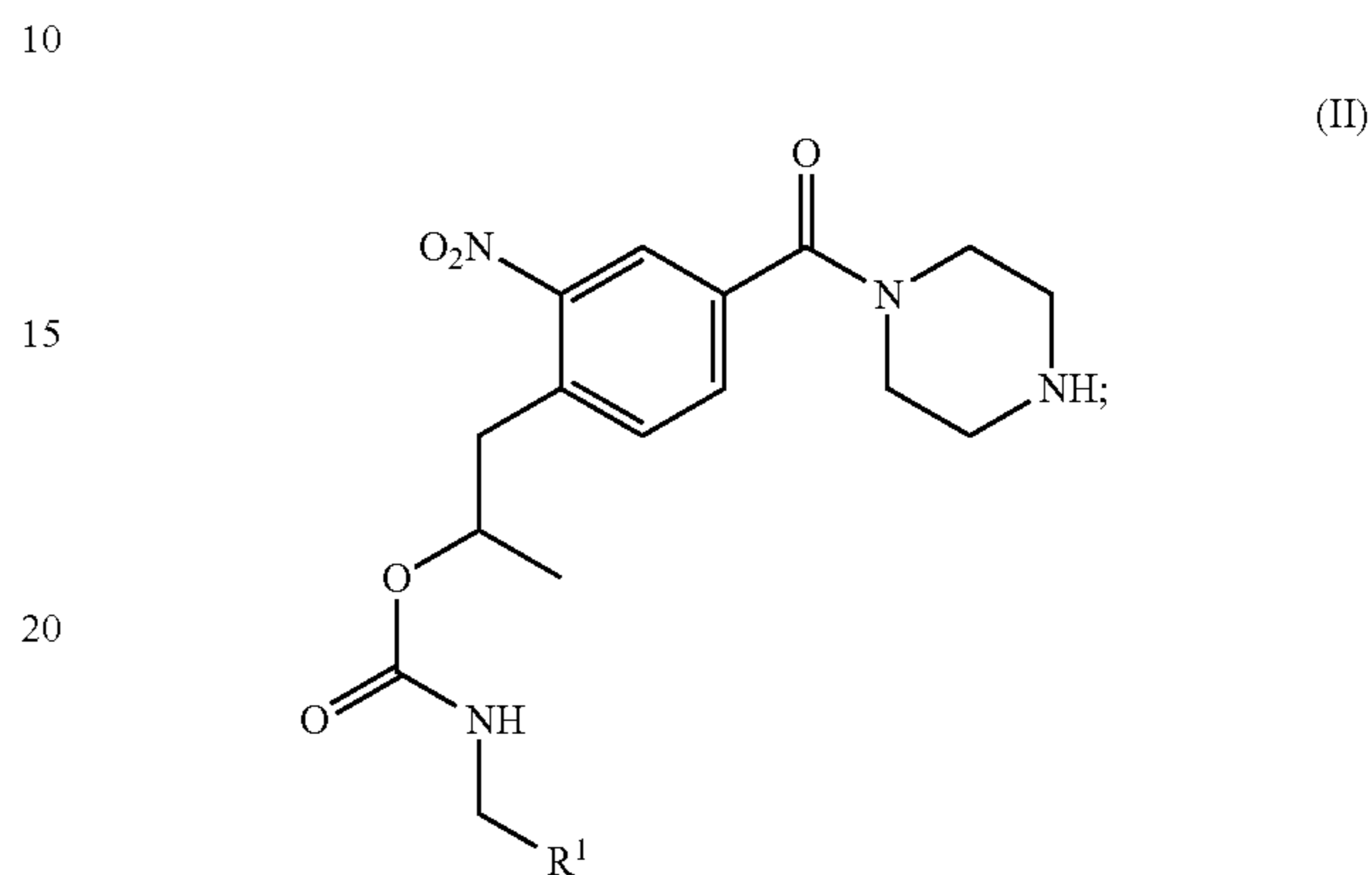
3. A compound of Formula (II-B):



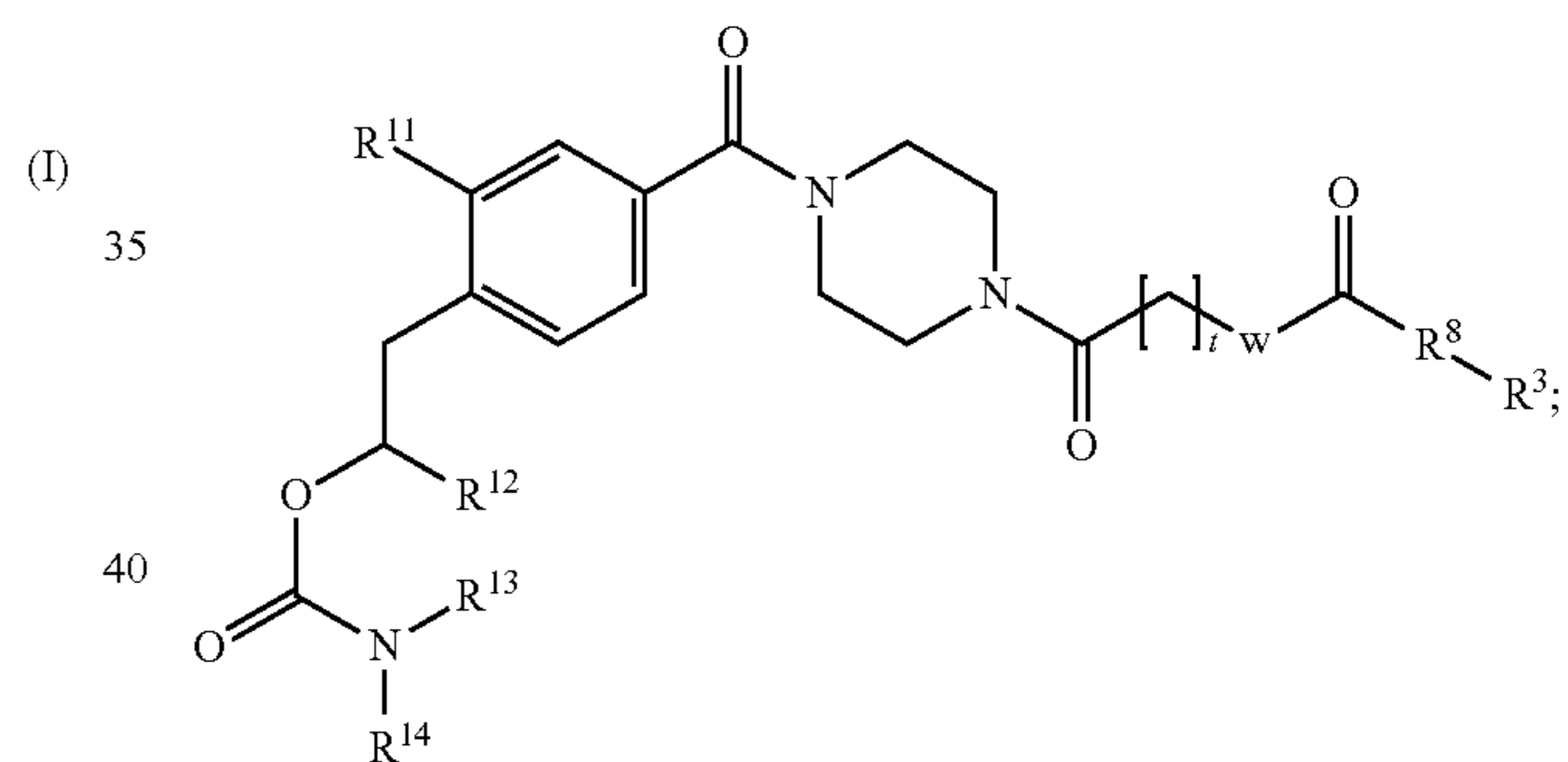
or a pharmaceutically acceptable salt thereof

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wherein

R¹¹ is selected from the group consisting of halo, CN,
and NO₂;R¹² is C₁₋₆-alkyl;R¹³ is H or C₁₋₆-alkyl; andR¹⁴ is a first linker bound to a support-medium.4. The compound of claim 3, wherein the compound of
Formula (II-B) is Formula (II):wherein R¹ is a support-medium.

5. A compound of Formula (B3):

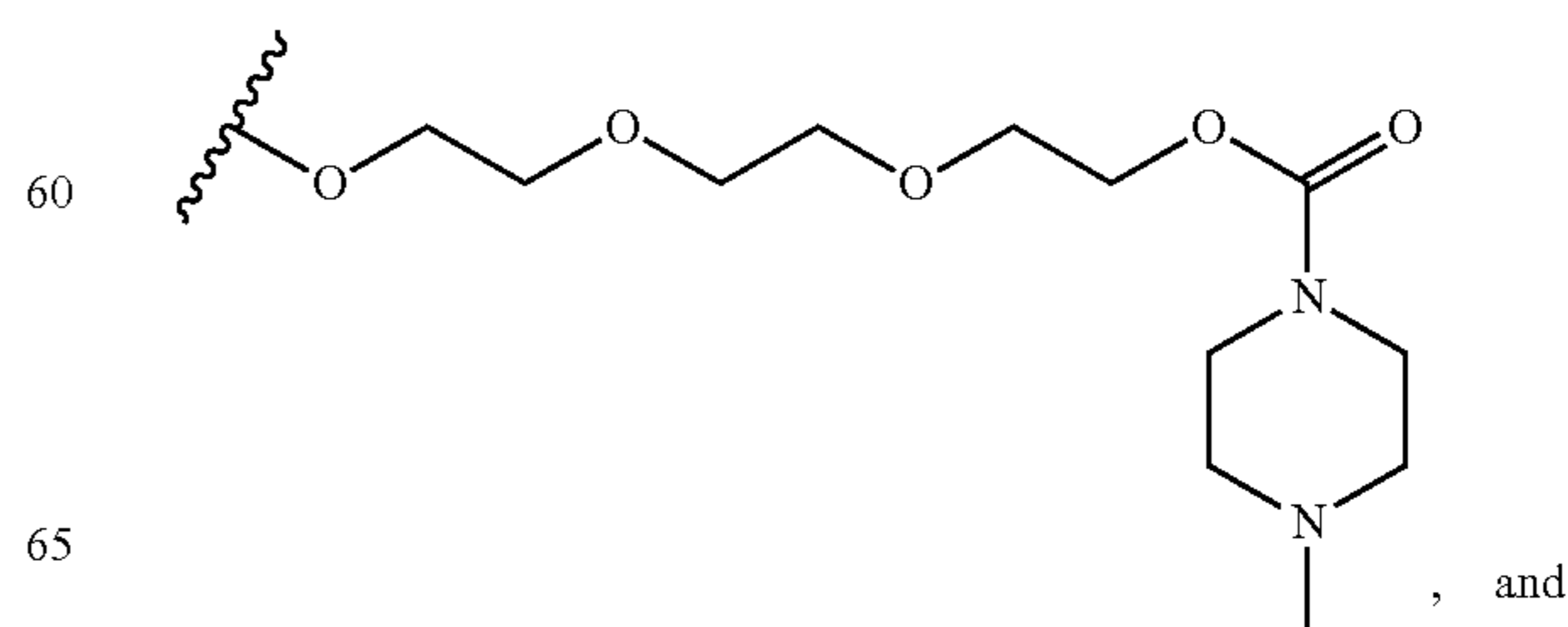


or a pharmaceutically acceptable salt thereof

wherein

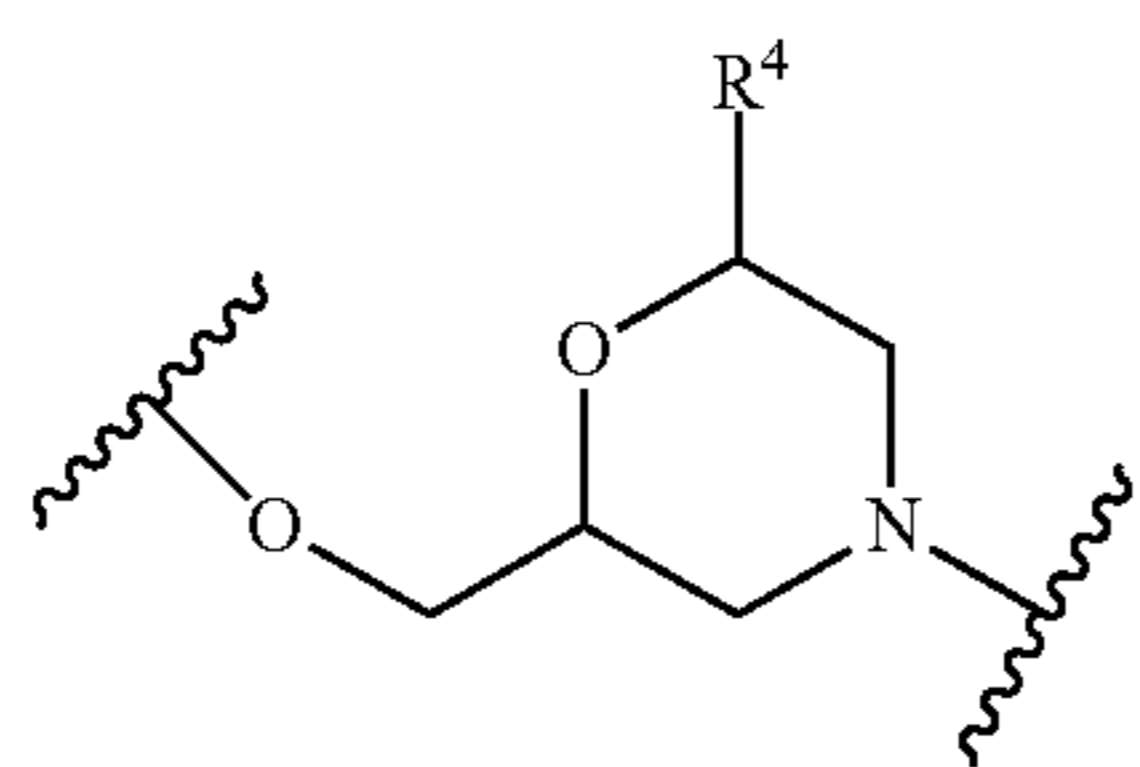
w is selected from O or C;

t is 1, 2, 3, 4, or 5;

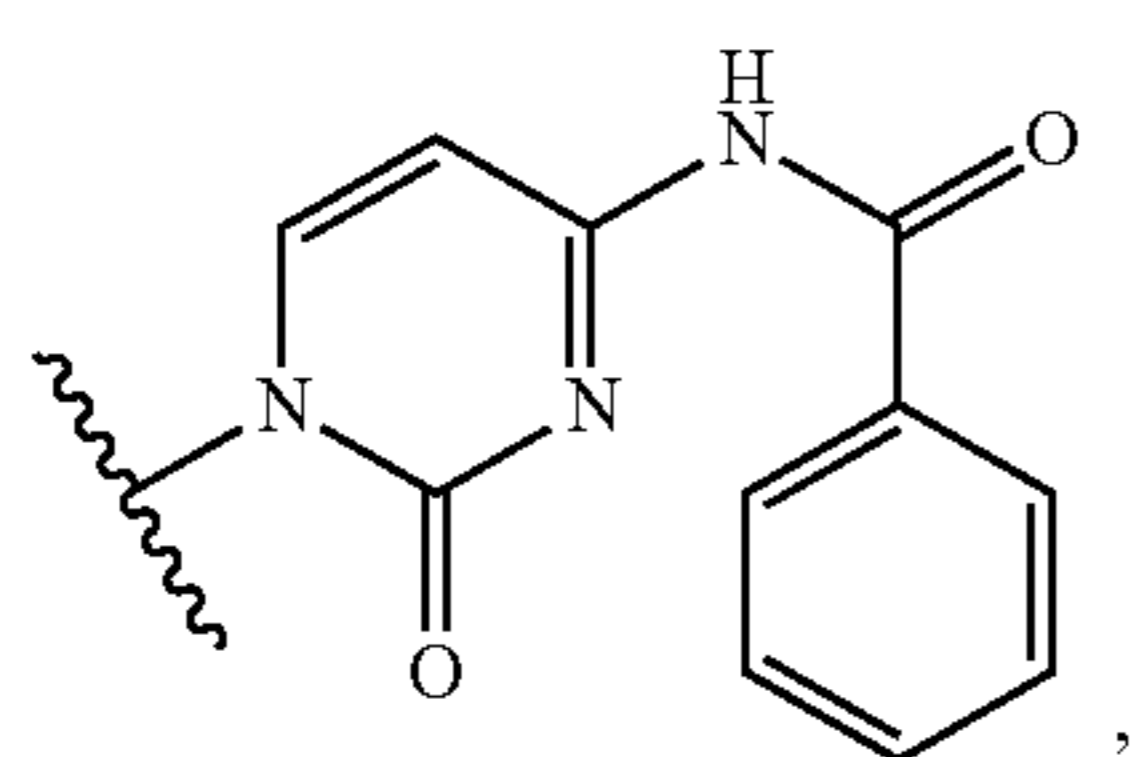
R¹¹ is selected from the group consisting of halo, CN,
and NO₂;R¹² is C₁₋₆-alkyl;R¹³ is H or C₁₋₆-alkyl;R¹⁴ is a first linker bound to a support-medium;R⁸ is selected from:

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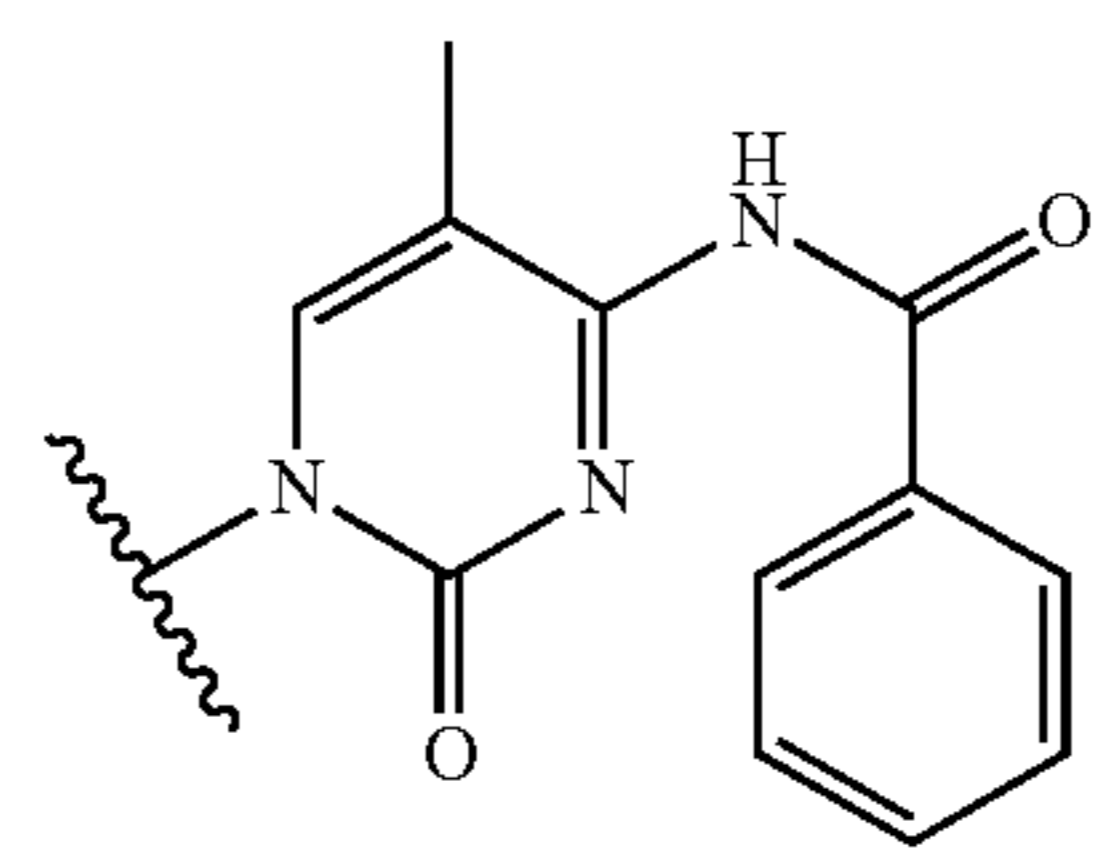
wherein R⁴ is selected from the group consisting of:



(PC) 15

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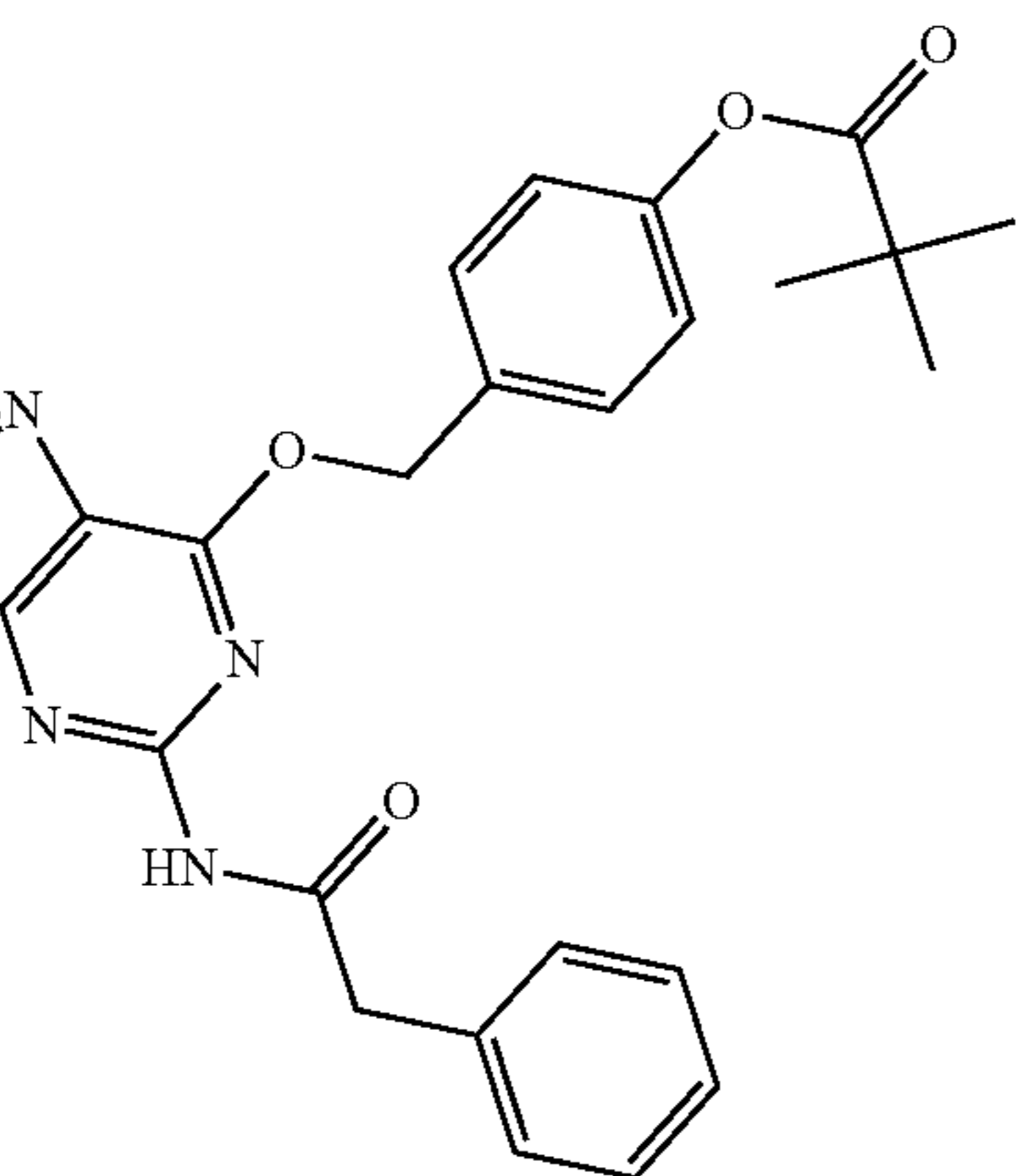
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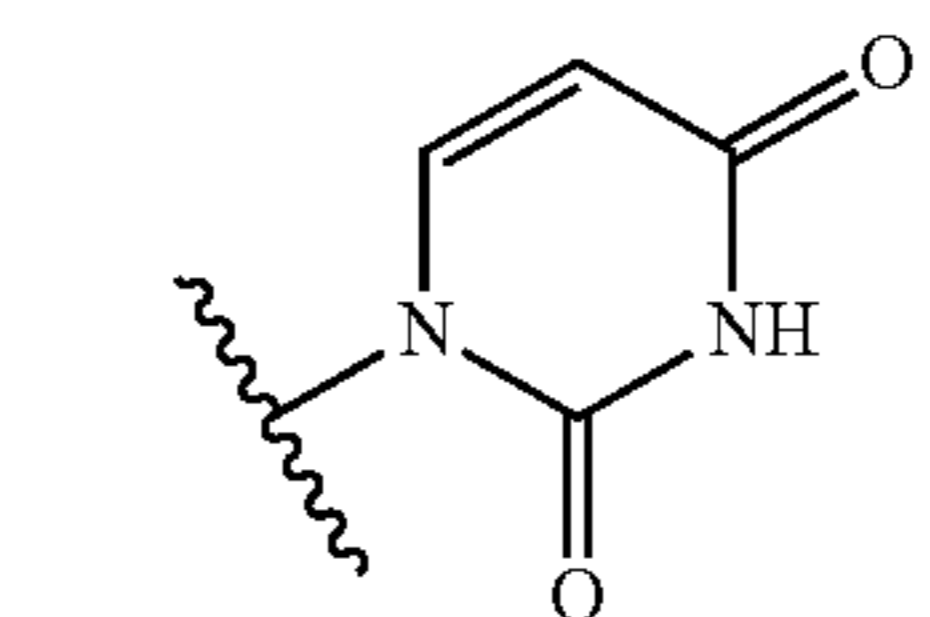


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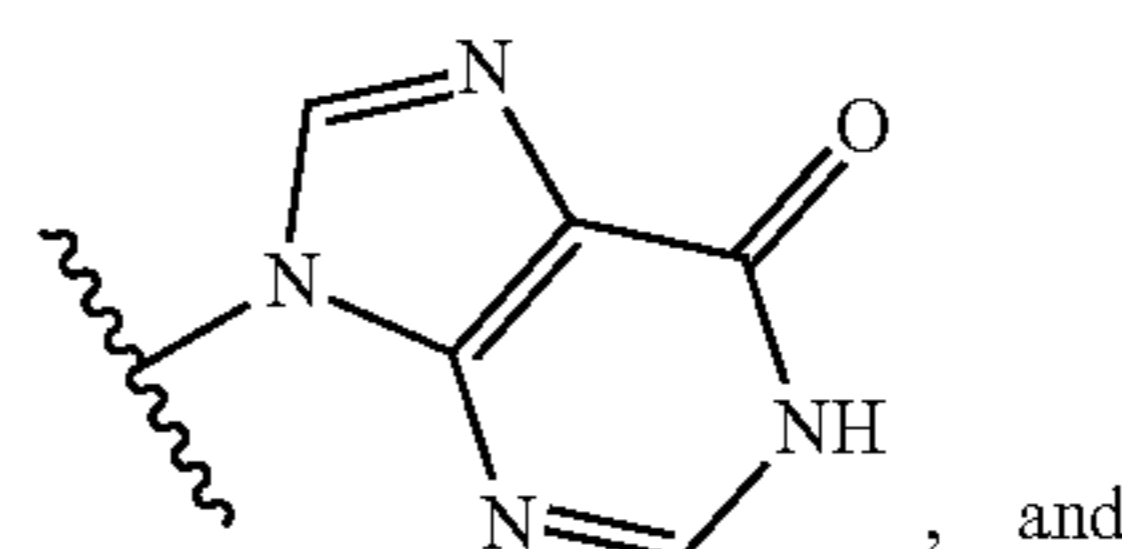
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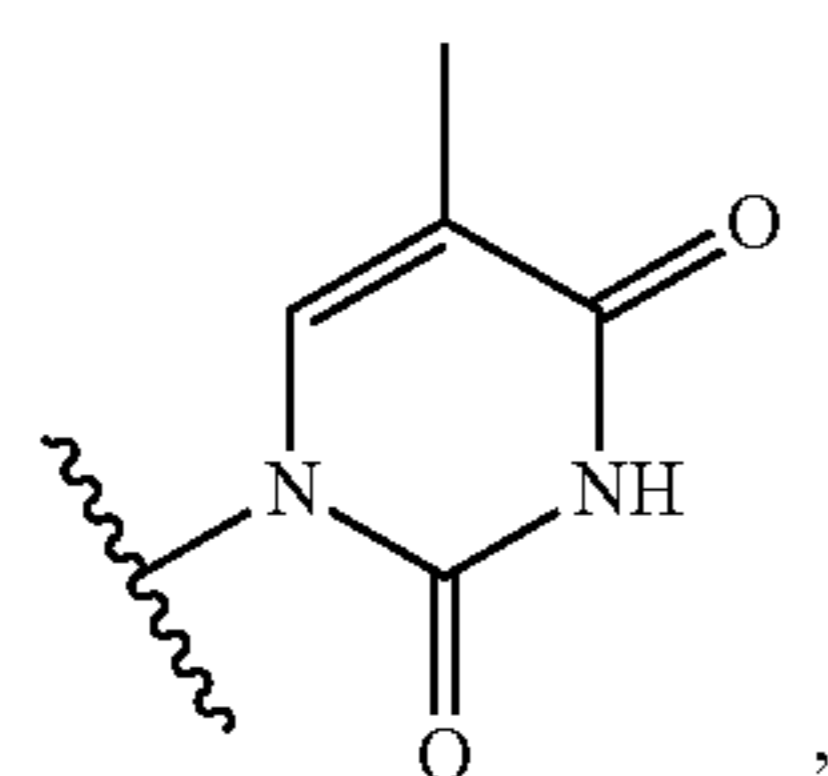
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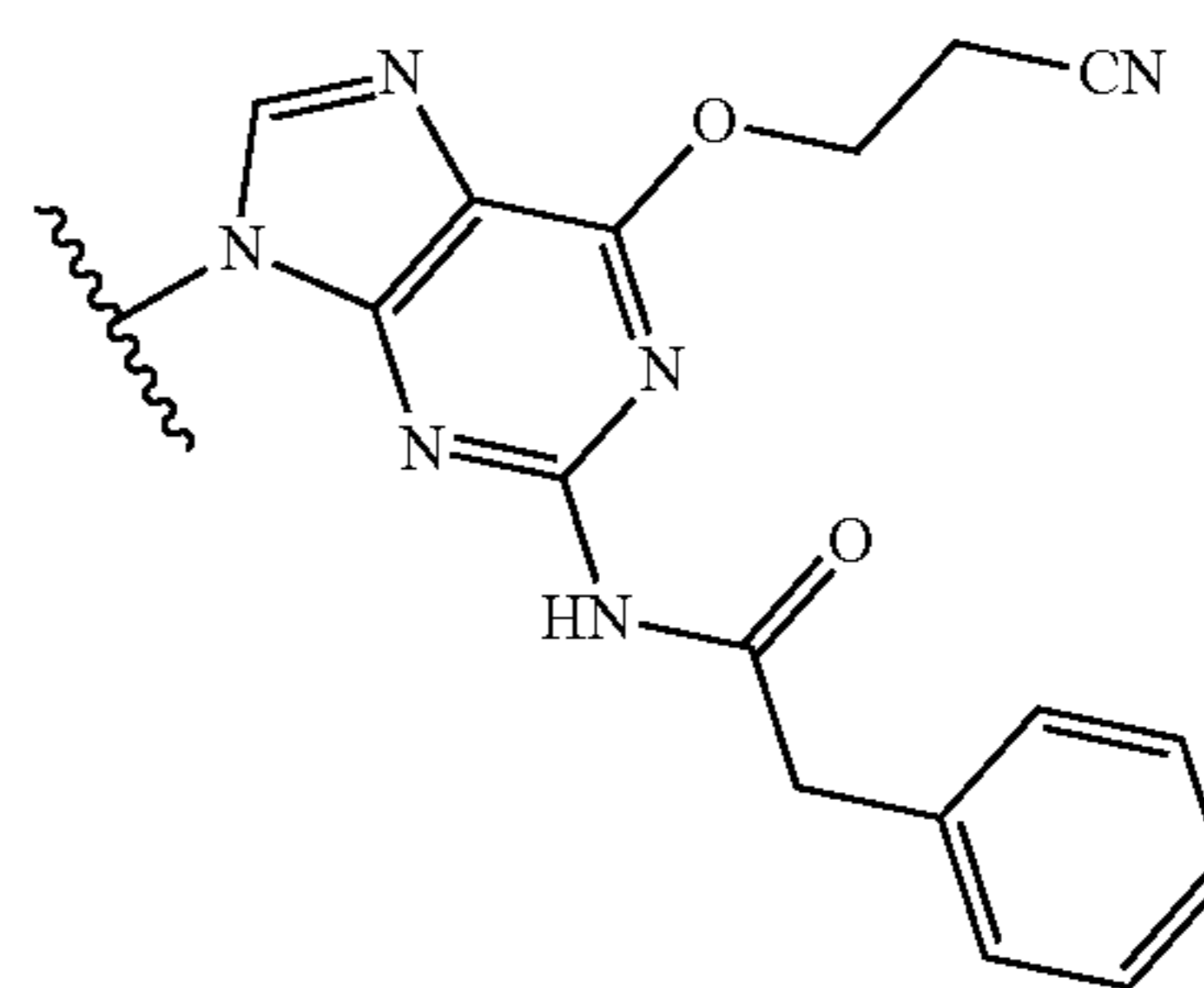


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(PG)

(PA)

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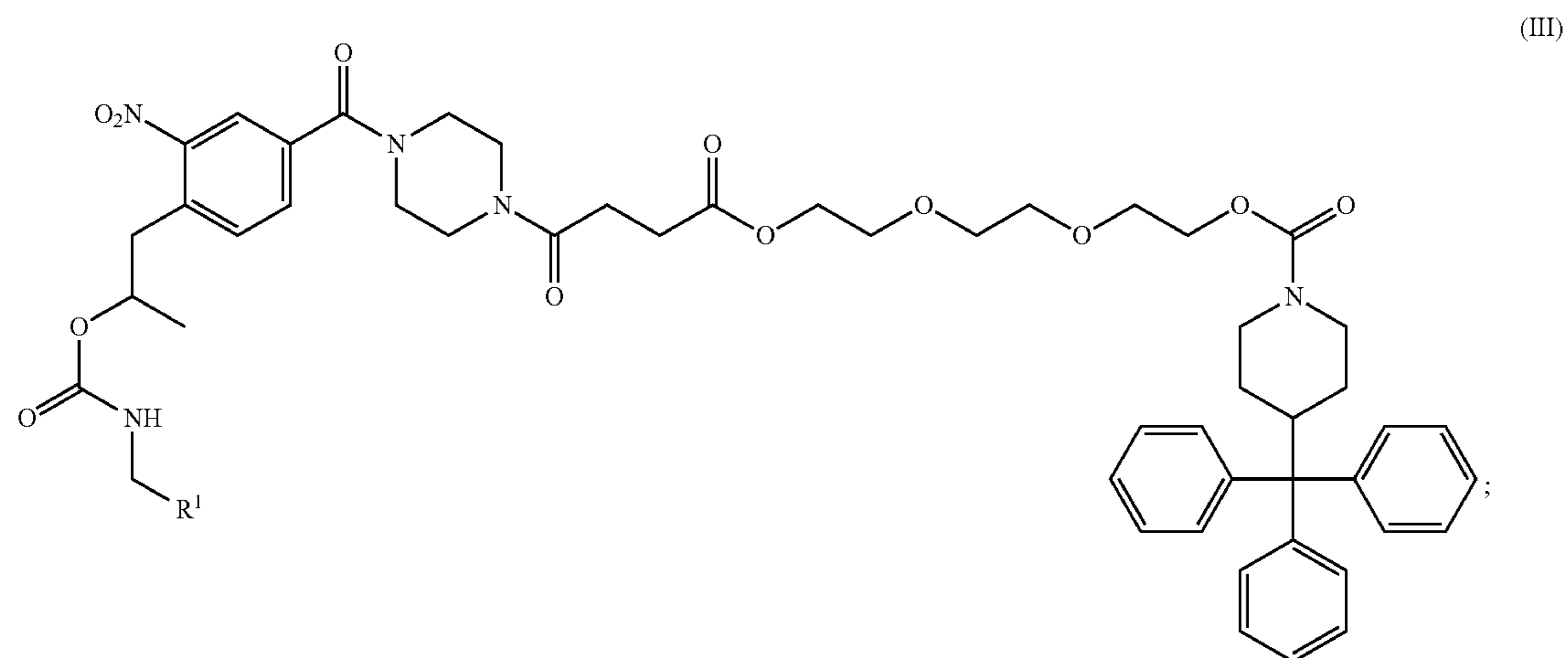
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65 R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl.

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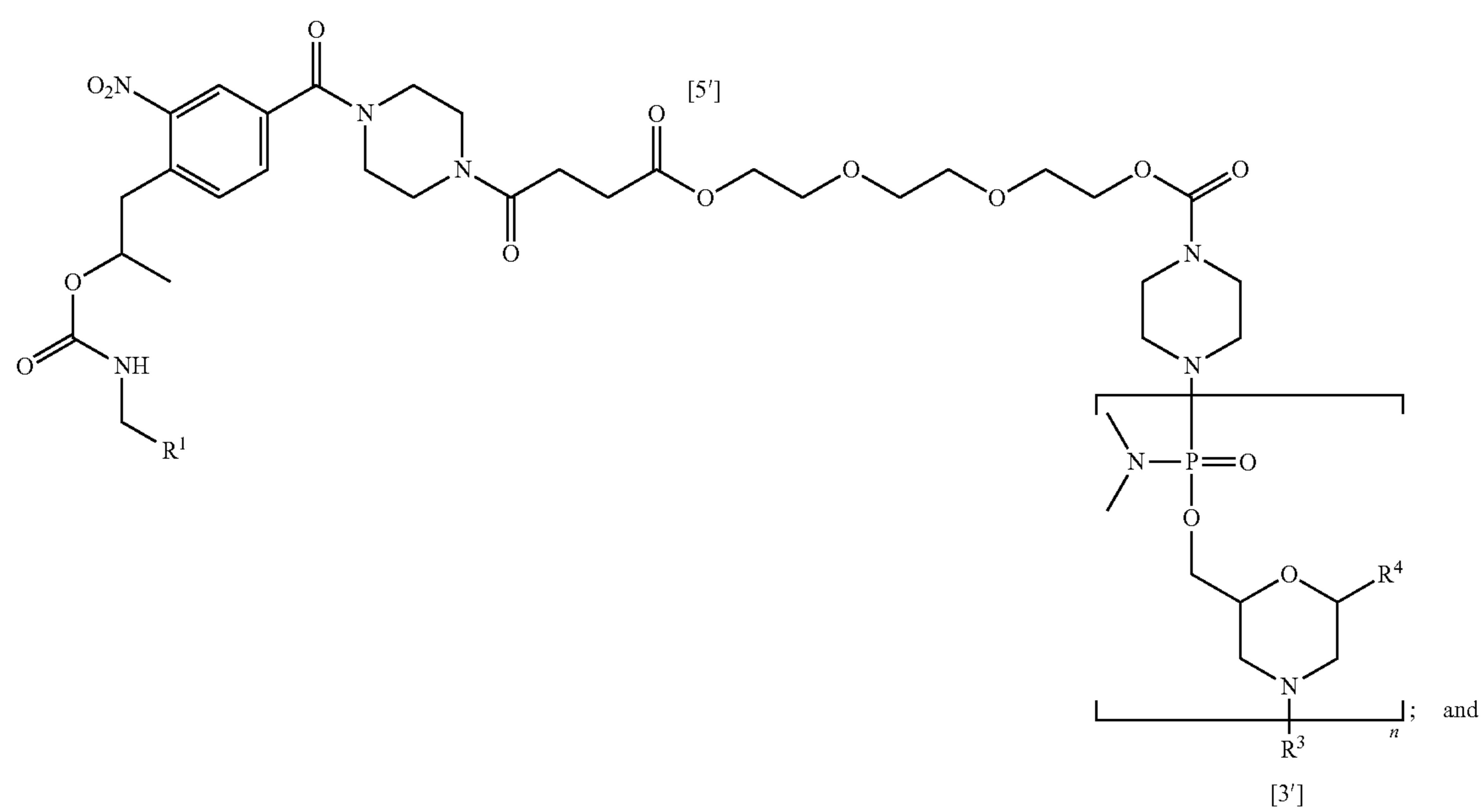
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6. The compound of claim 5, wherein the compound of Formula (B3) is of Formula (III):



wherein R¹ is a support-medium.
7. A compound selected from:

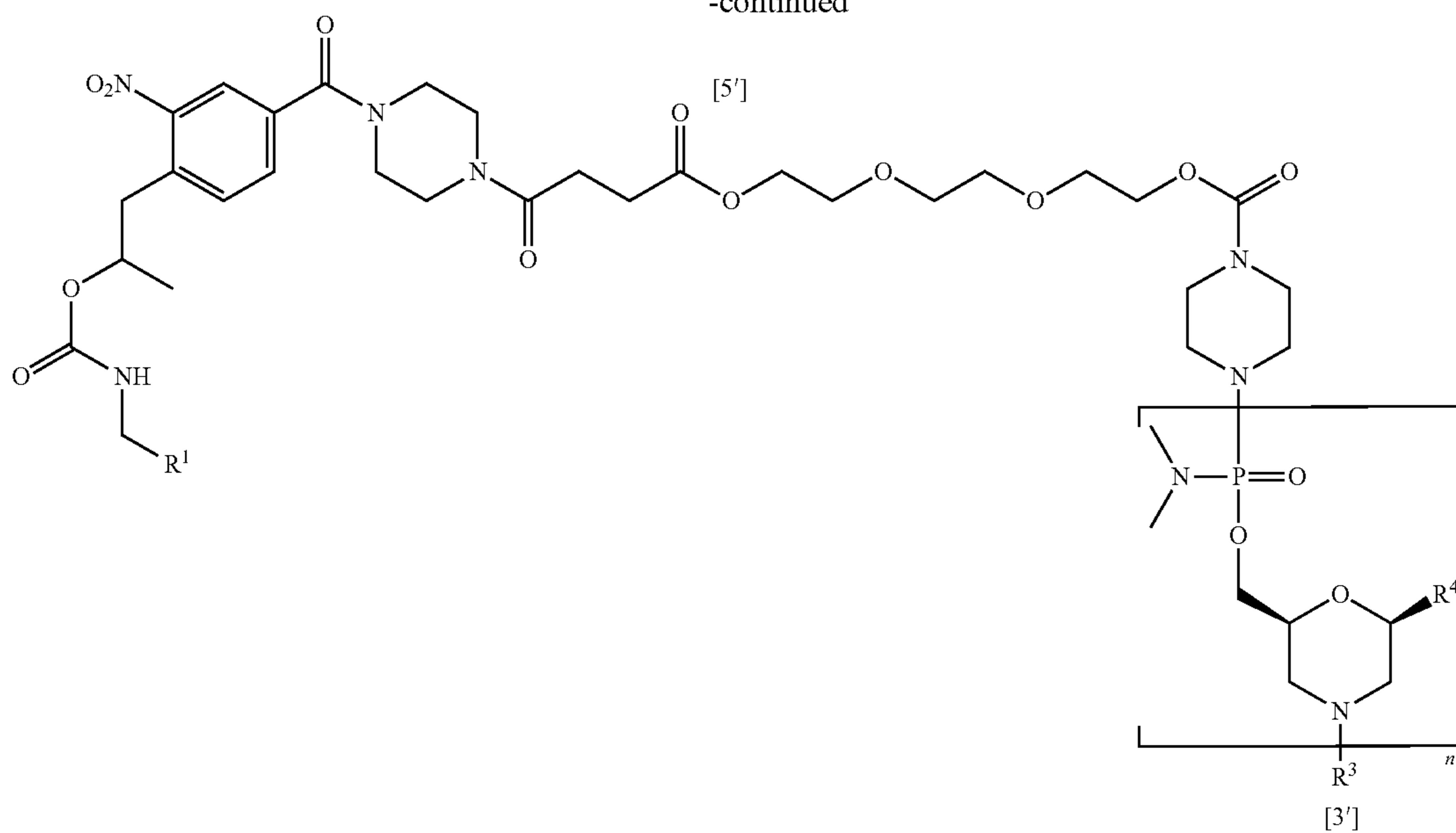
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wherein:

n is an integer from 10 to 40;

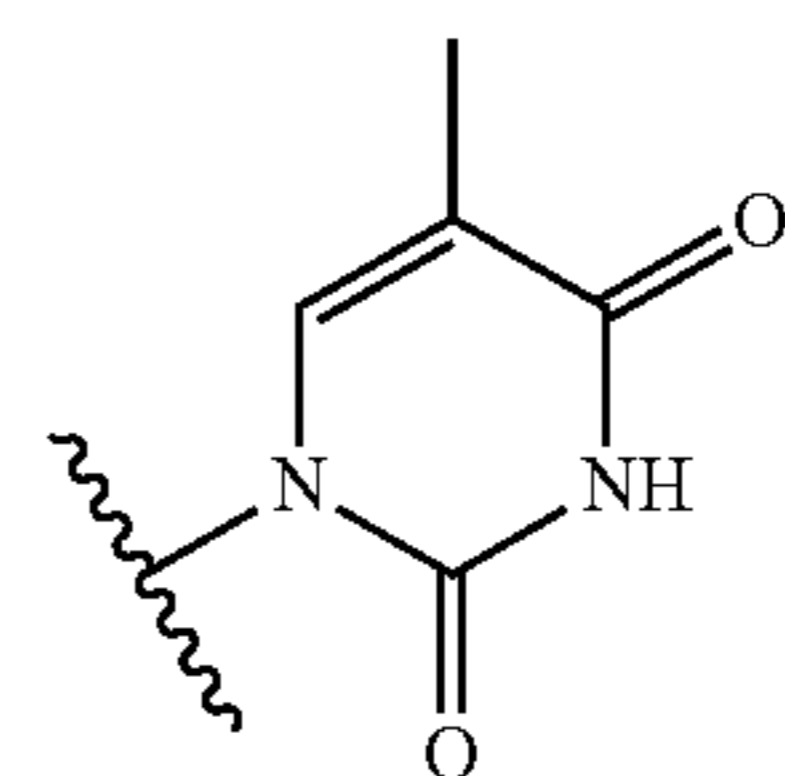
R¹ is a support-medium;

R³ is selected from the group consisting of trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl; and

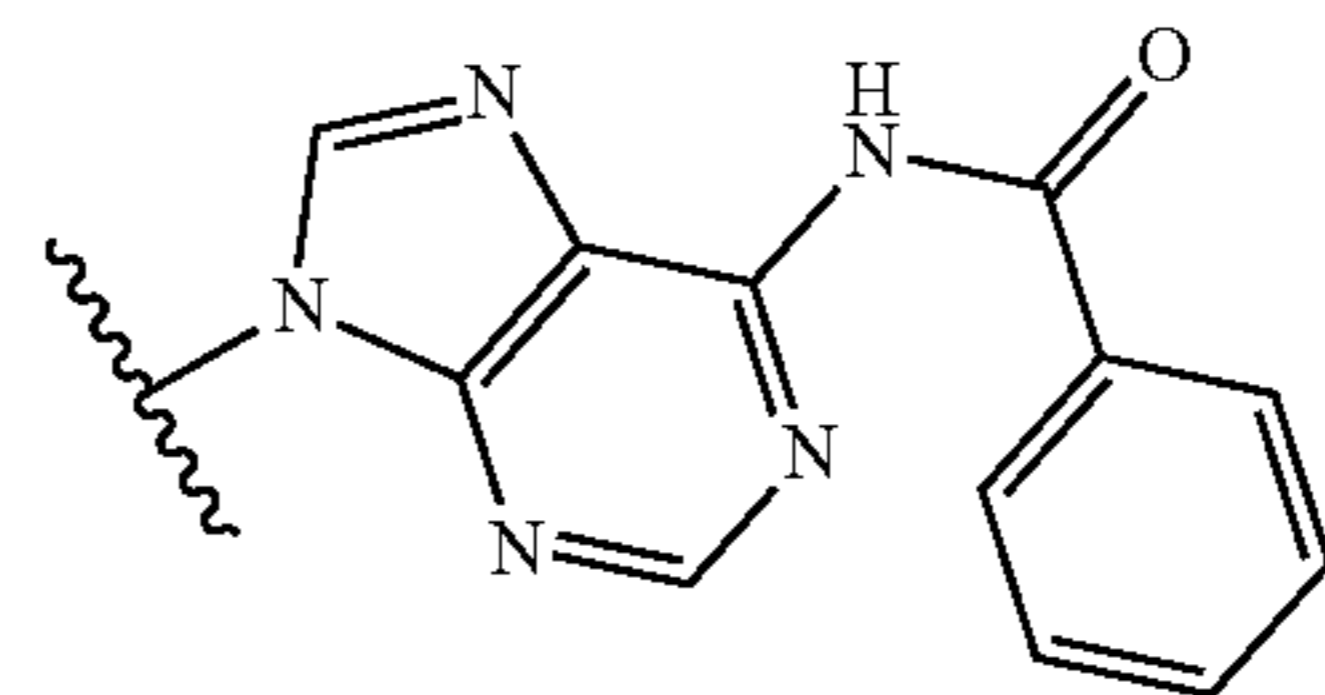
R⁴ is, independently at each occurrence, selected from the group consisting of:

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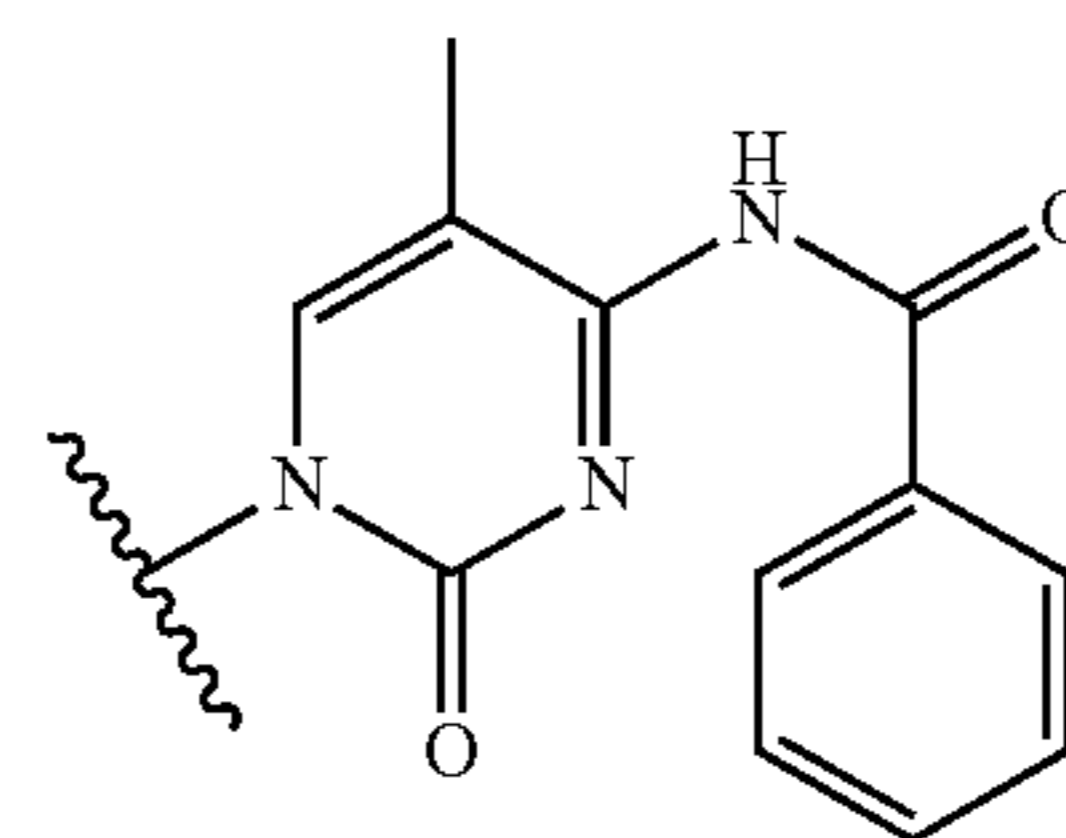
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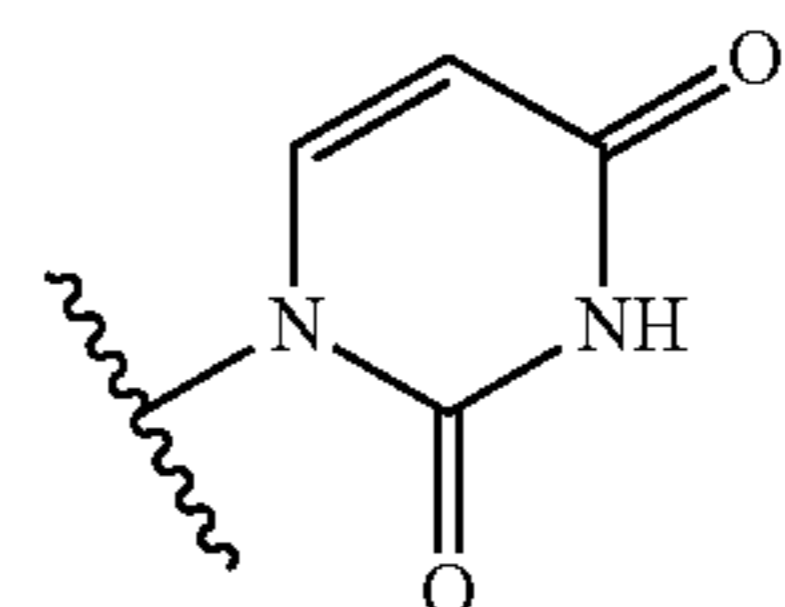
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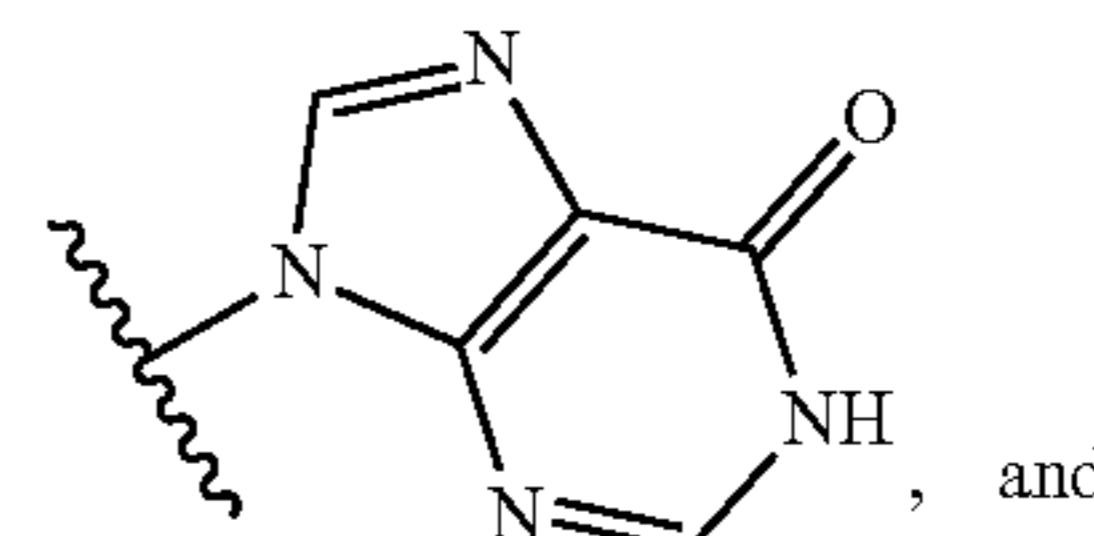
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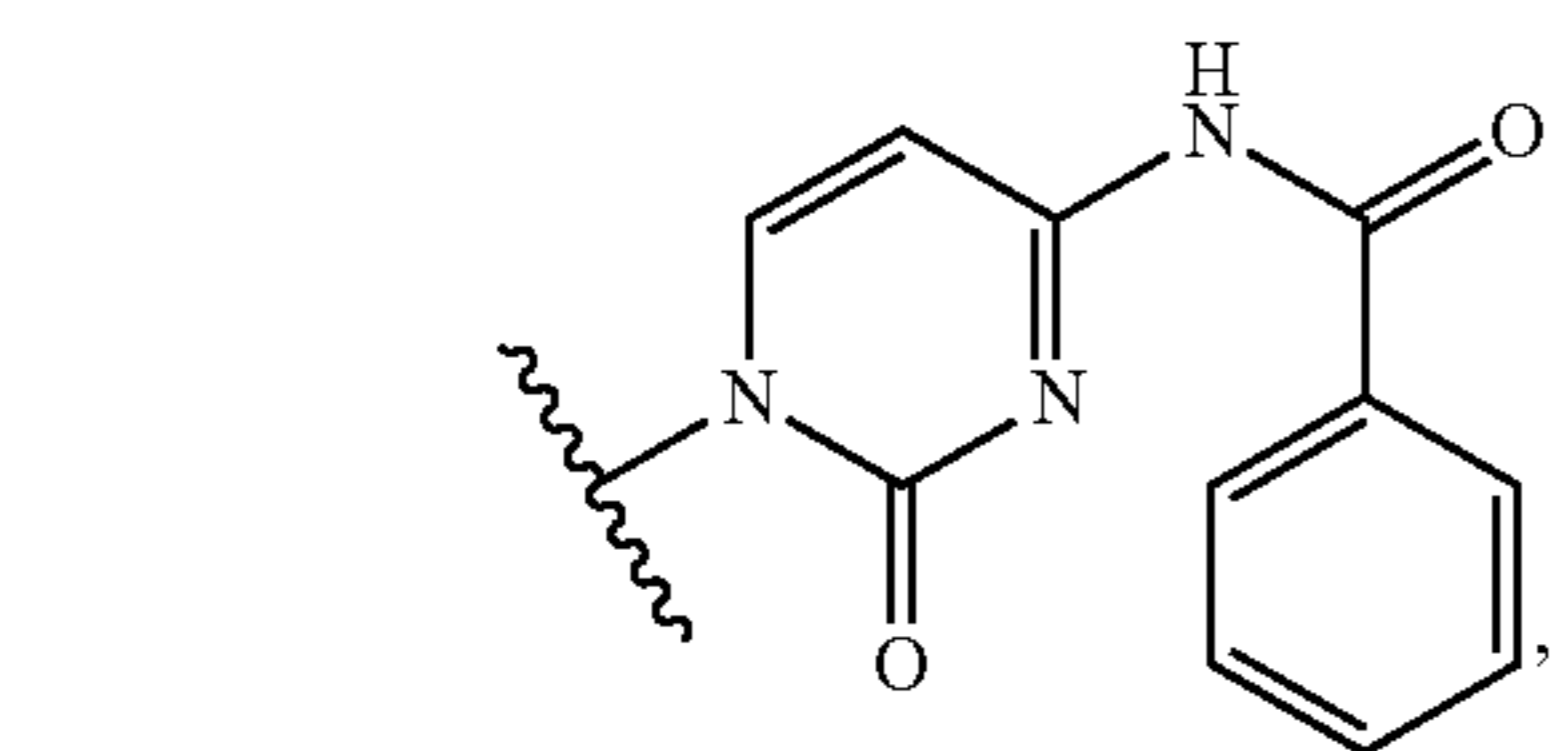
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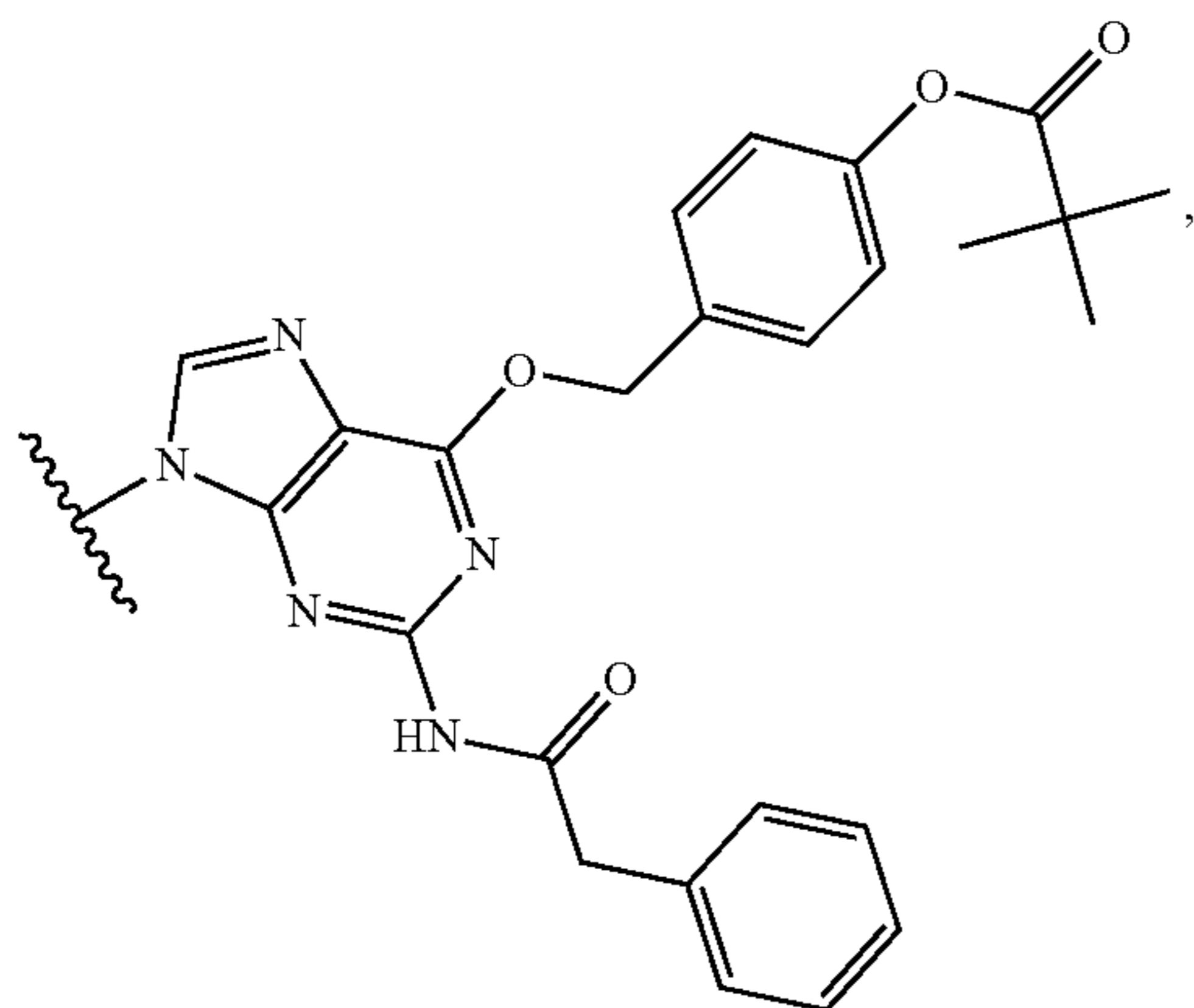


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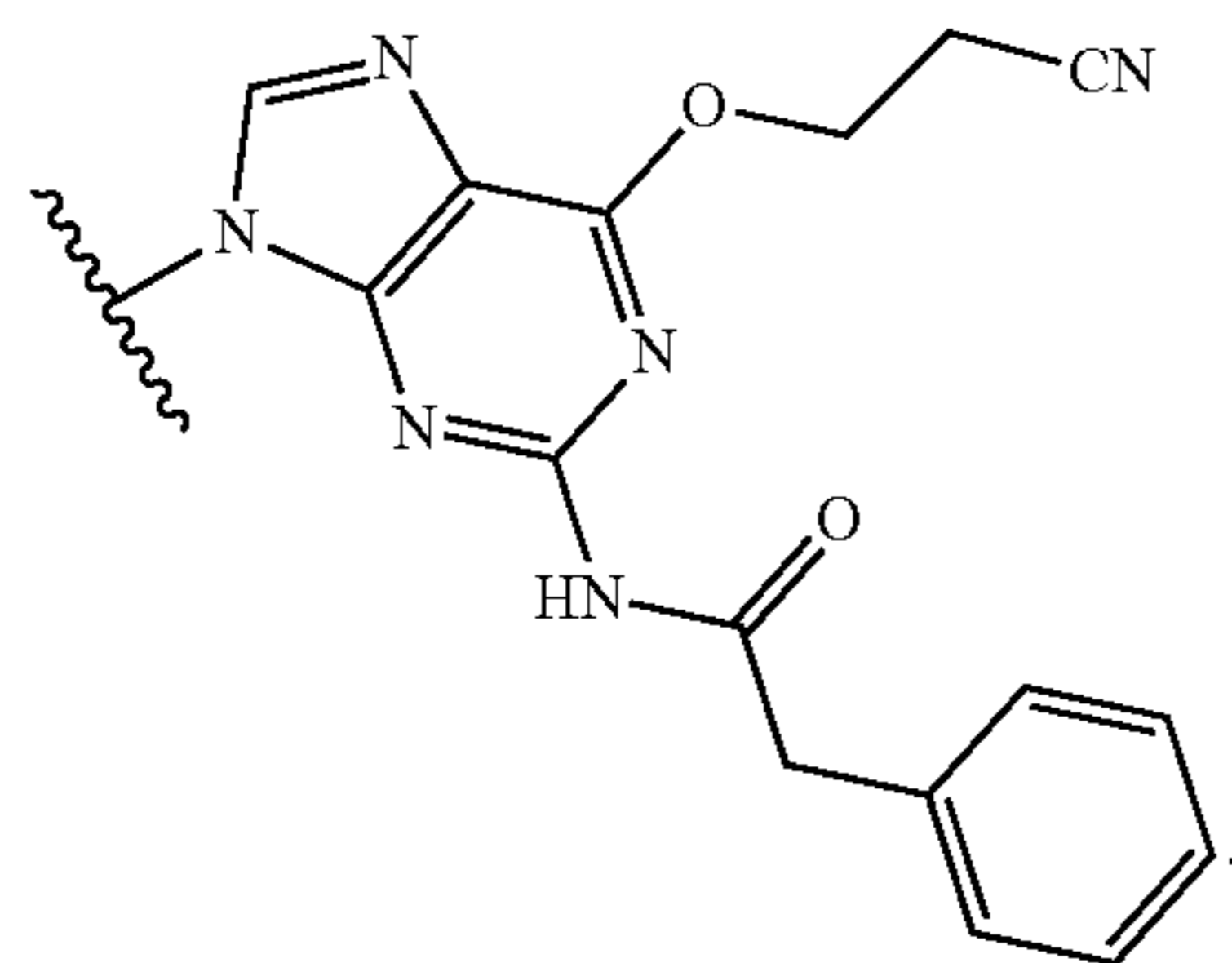
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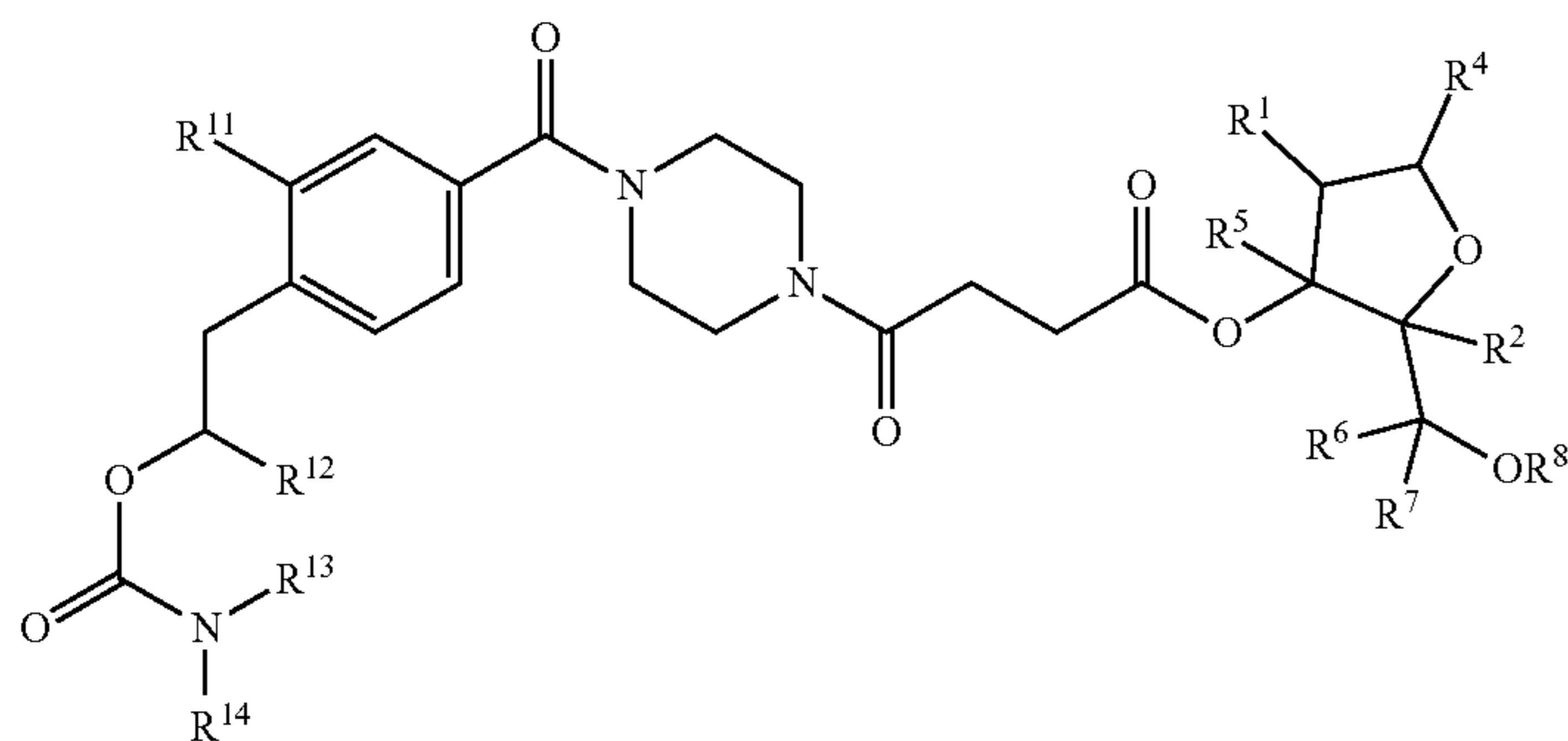


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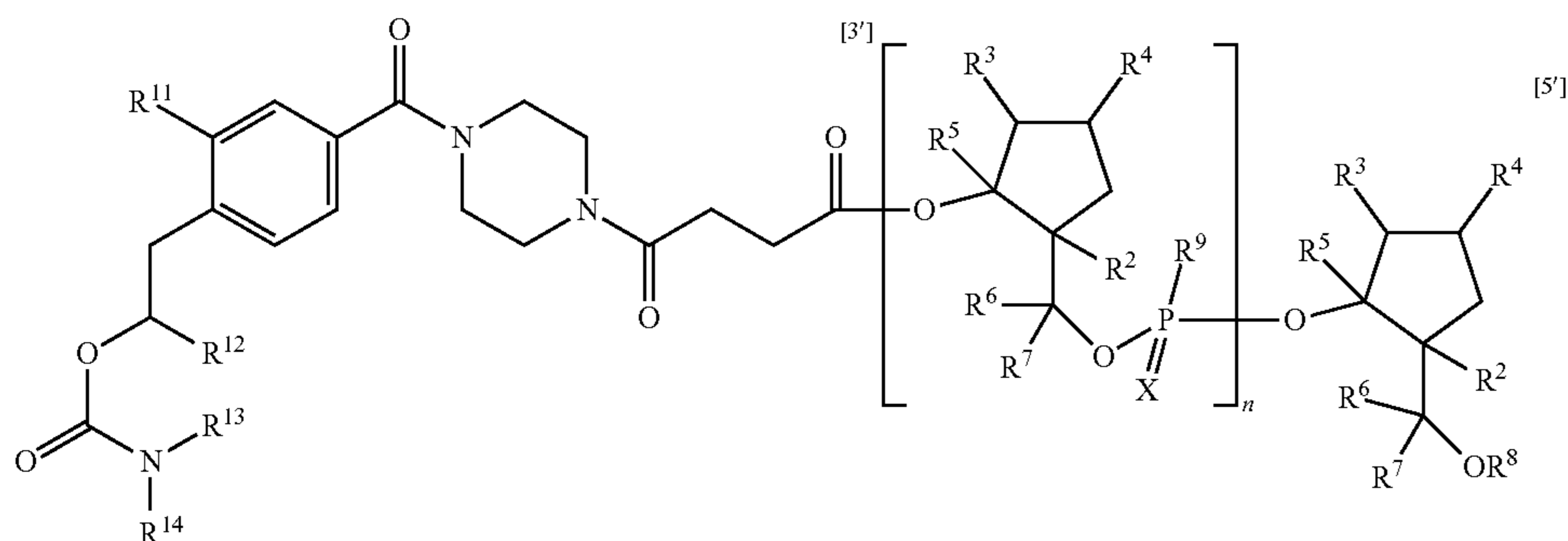
8. A compound selected from:



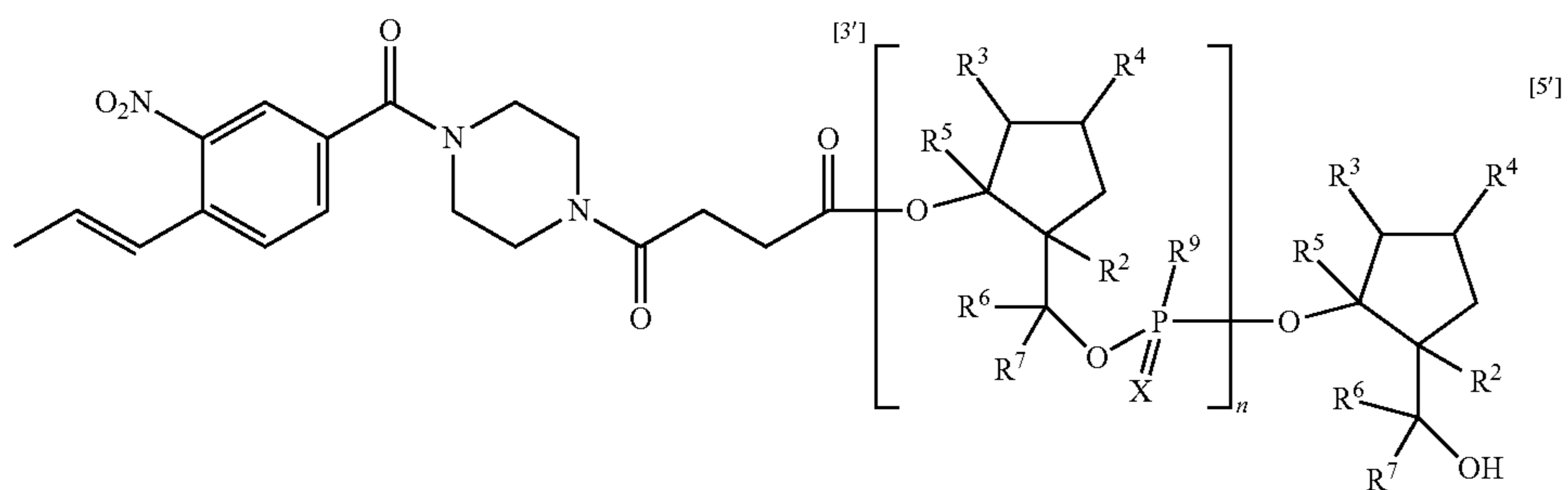
R^2 is H or R^1 and R^2 join to form a cycloalkyl or heterocyclic ring having from 4 to 7 ring atoms optionally containing an oxygen;

R^5 , R^6 , and R^7 are each hydrogen, or R^5 and R^7 join to form a cycloalkyl ring having from 4 to 7 ring atoms and R^6 and R^7 join to form a cycloalkyl ring having from 3 to 4 ring atoms,

R^9 is $-\text{OCH}_2\text{CH}_2\text{CN}$ or $-\text{CH}_2\text{C}(=\text{O})\text{OC}(\text{CH}_3)_2\text{CH}_2\text{CN}$;



; and



wherein:

R^{11} is selected from the group consisting of halo, CN, and NO_2 ;

R^{12} is C_{1-6} -alkyl;

R^{13} is H or C_{1-6} -alkyl;

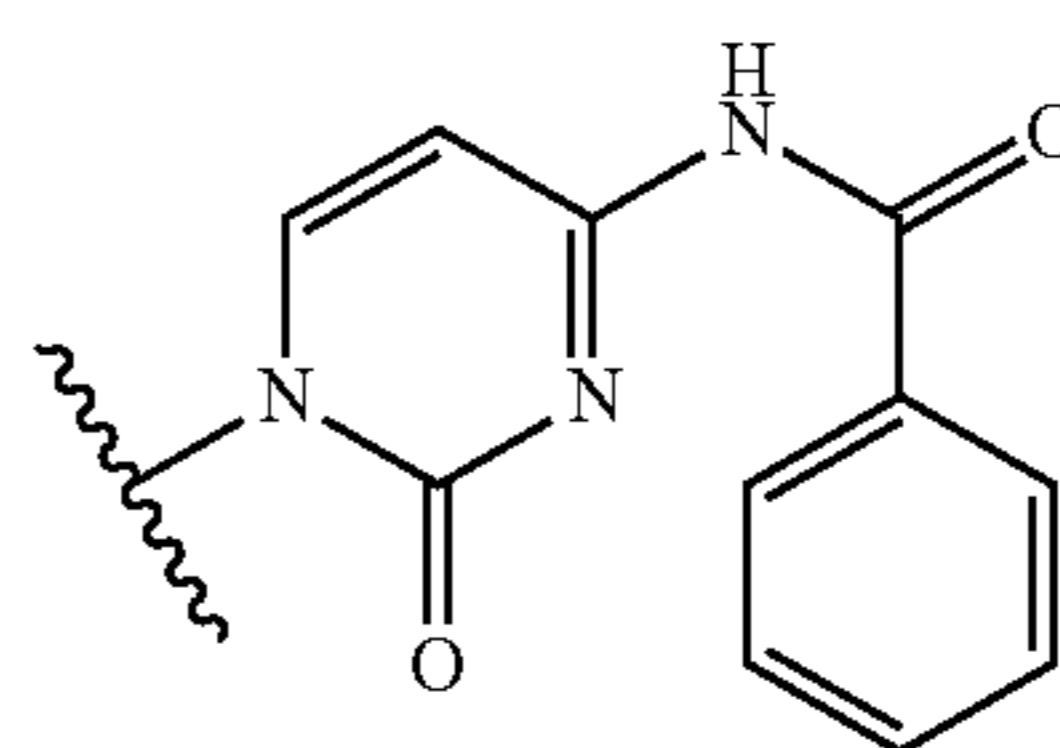
R^{14} is a first linker bound to a support-medium;

R^1 is selected from $-\text{O}(\text{C}_1\text{-C}_6\text{alkyl})$, halo, $-\text{O}(\text{C}_1\text{-C}_6\text{alkyl})-\text{O}(\text{C}_1\text{-C}_6\text{alkyl})$, and $-\text{O}(\text{C}_1\text{-C}_6\text{alkyl})-\text{C}(\text{O})-\text{NH}-\text{C}_1\text{-C}_6\text{alkyl}$

55

R^4 is, independently at each occurrence, selected from the group consisting of:

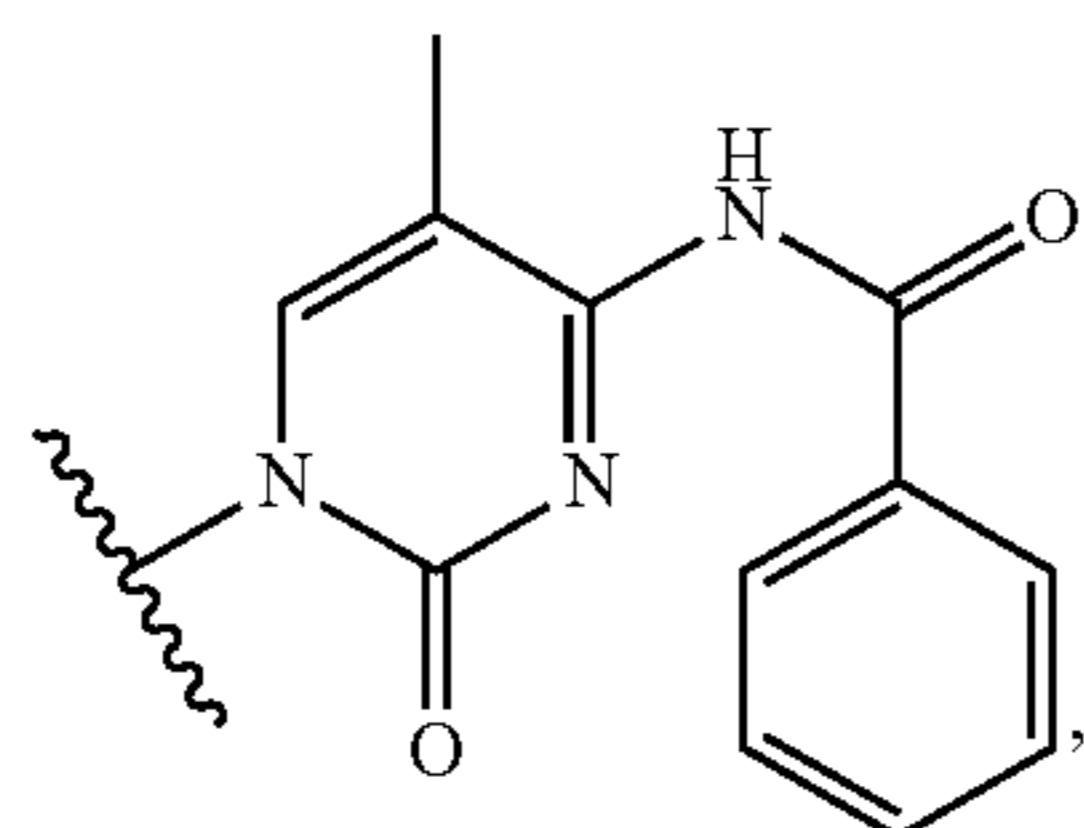
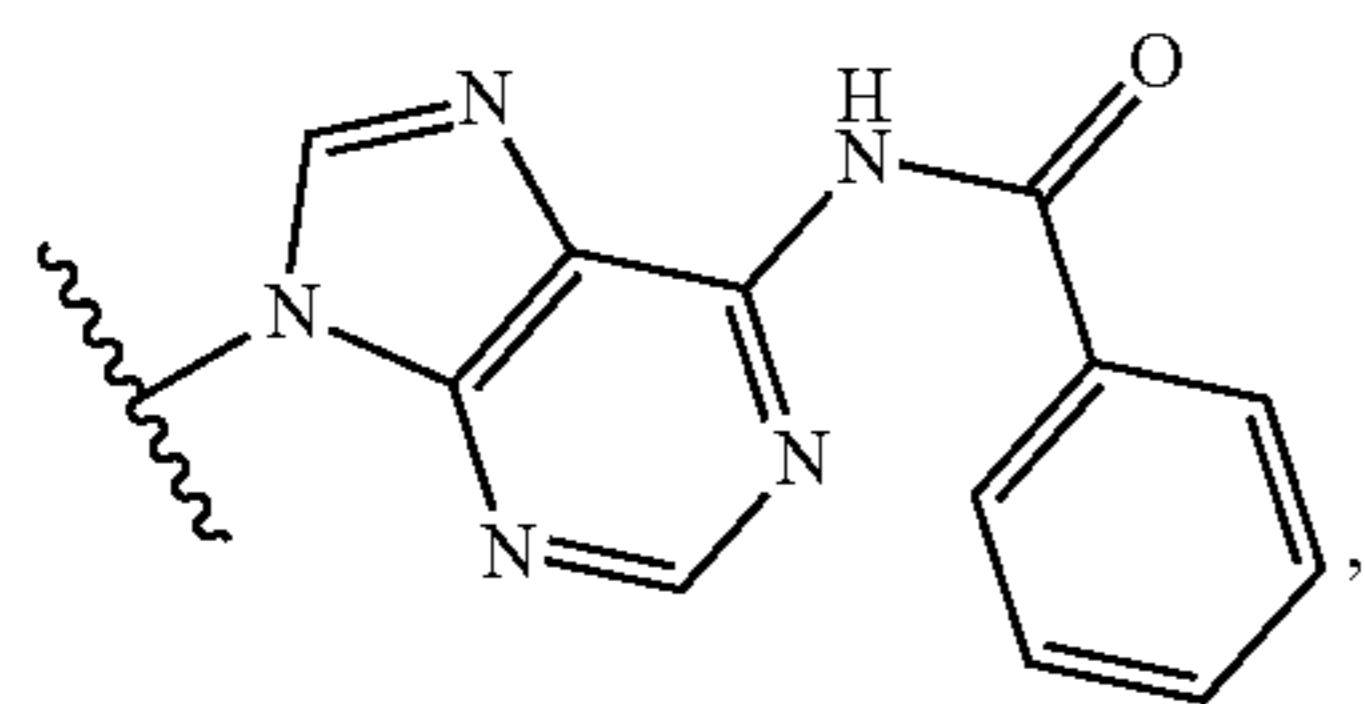
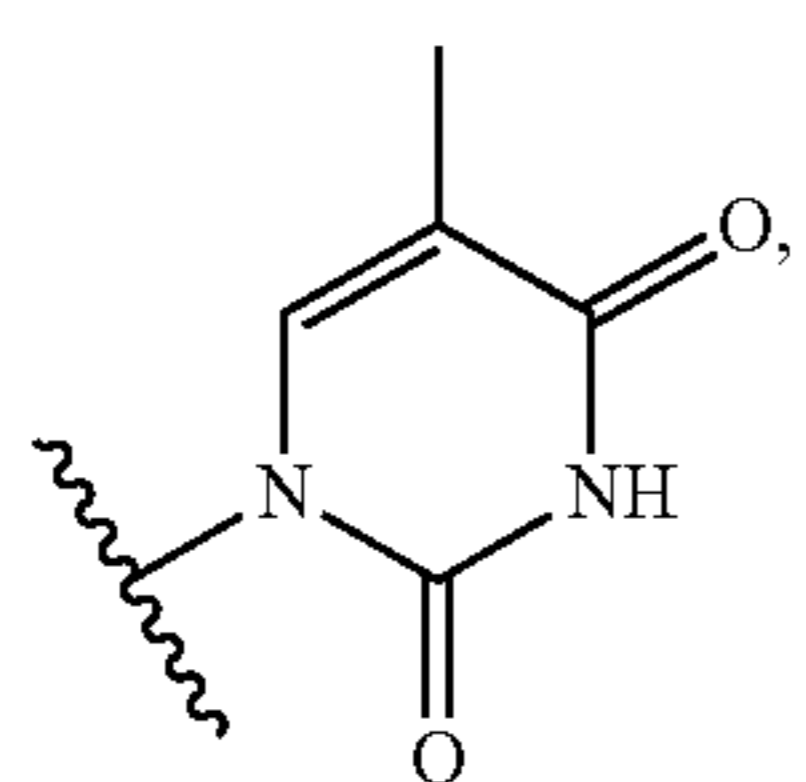
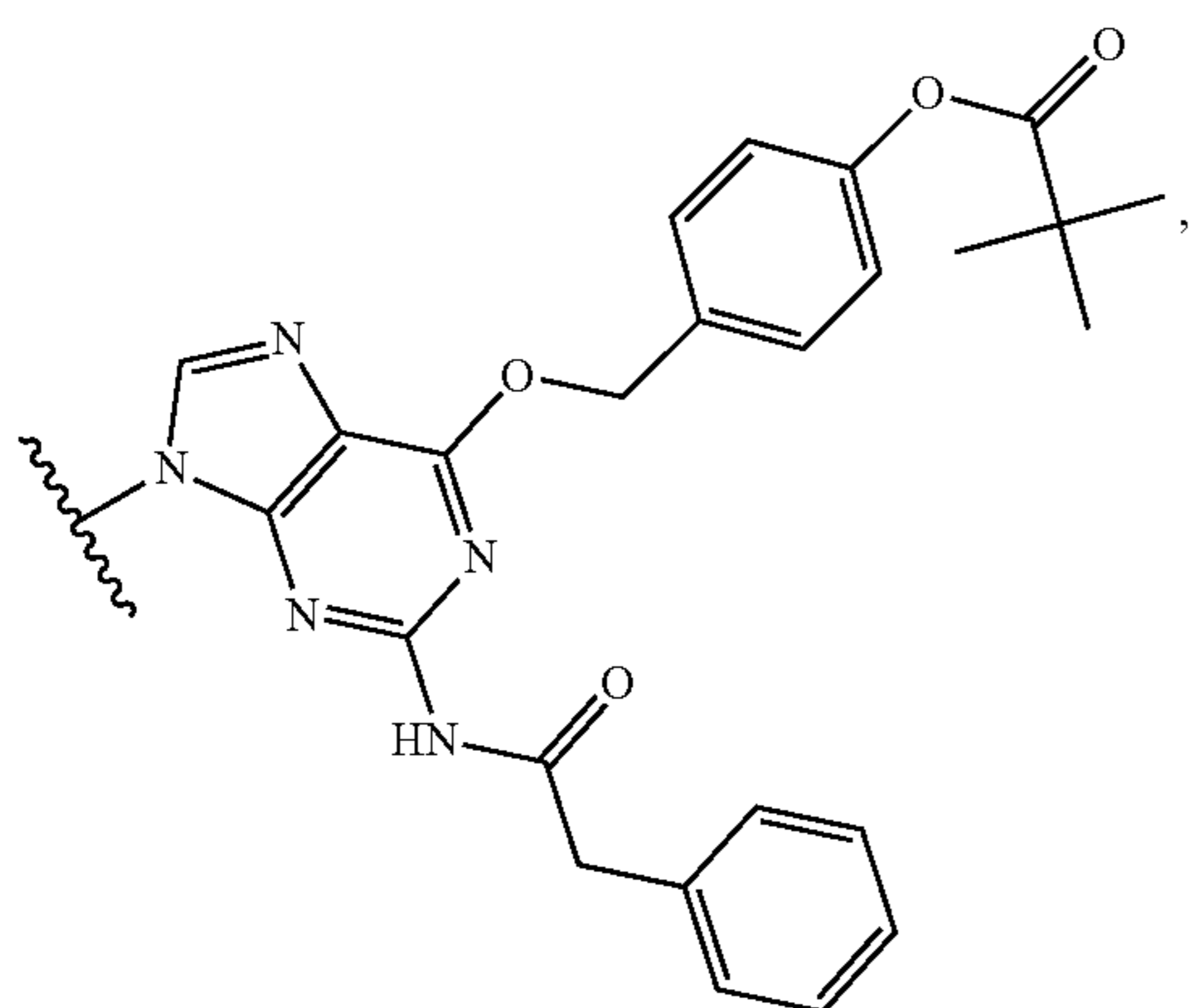
60



(PC)

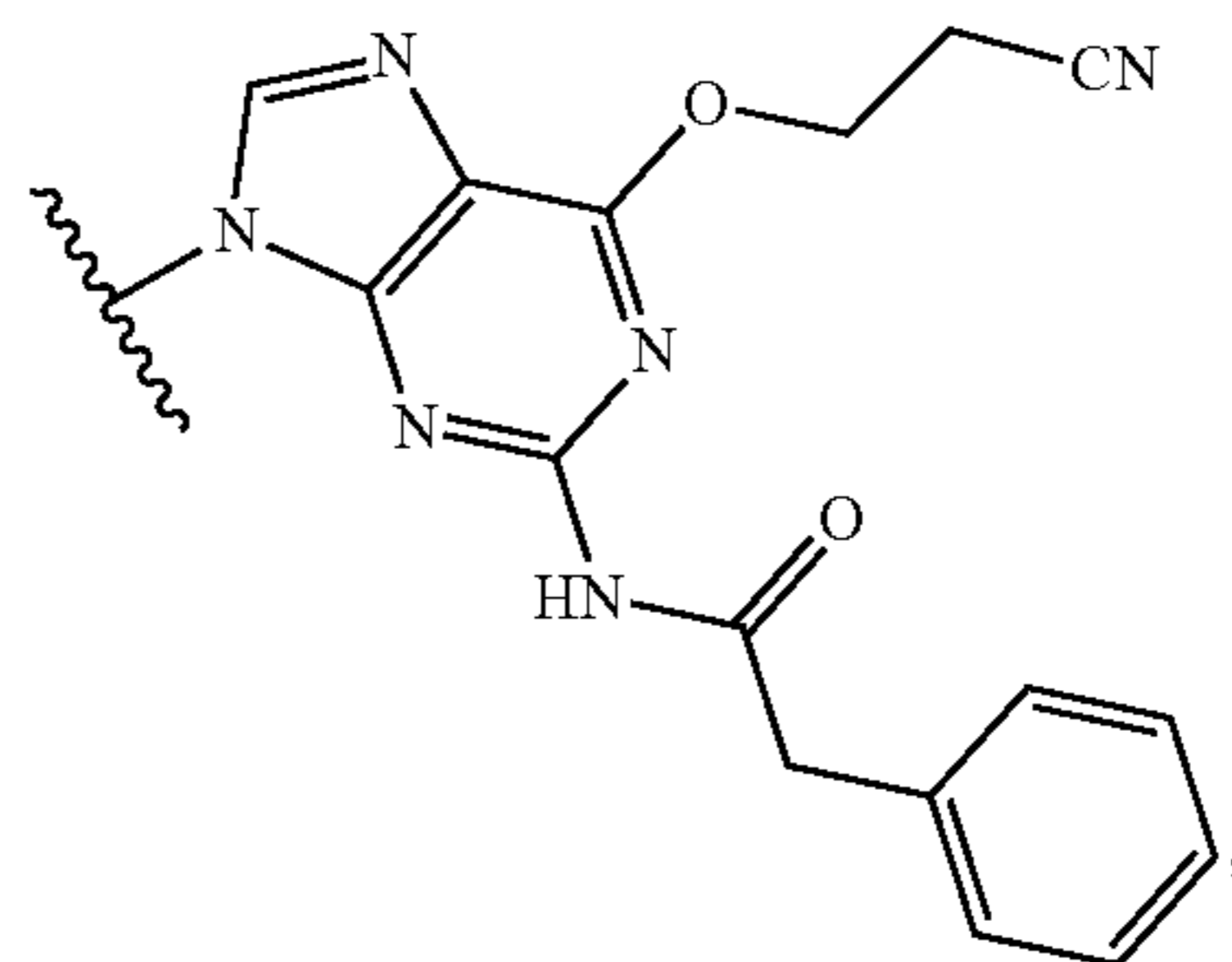
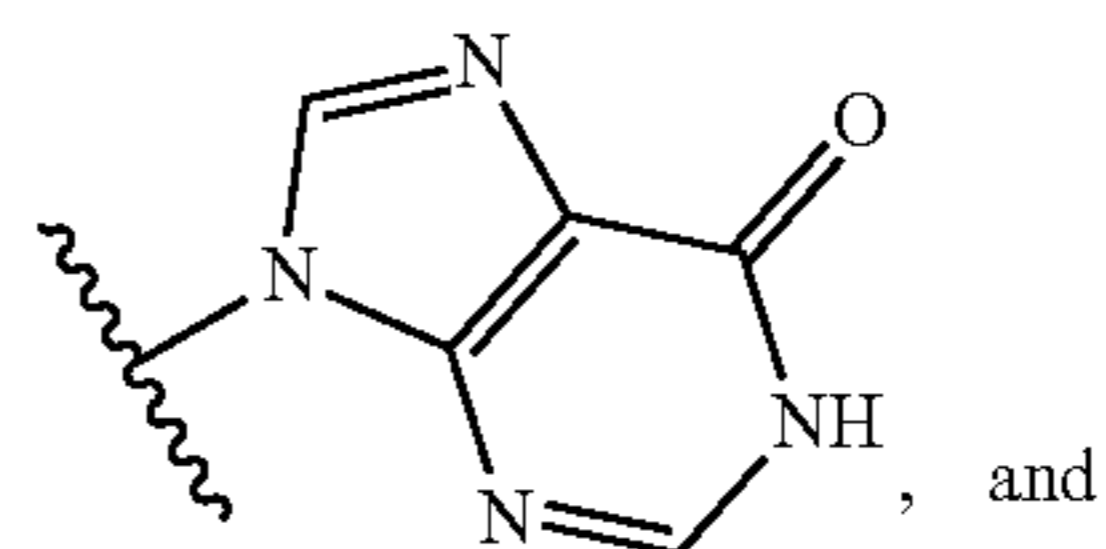
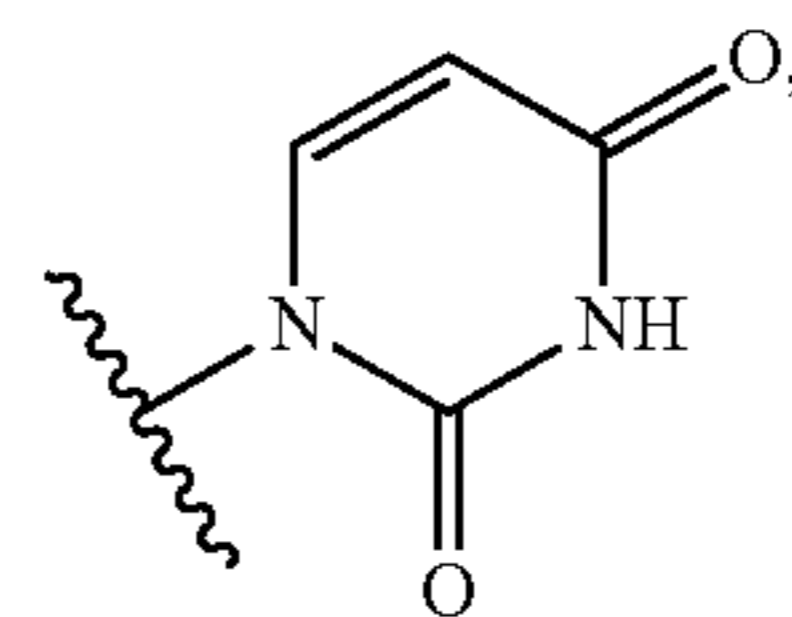
283

-continued



284

-continued



R⁸ is, independently at each occurrence, selected from the group consisting of H, trityl, monomethoxytrityl, dimethoxytrityl and trimethoxytrityl; n is 10 to 40; and X is O or S.

* * * * *